CONF-710809

TRITIUM

A. Alan Moghissi, et al

University of Nevada
Las Vegas, Nevada

May 1973

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Phoenix, Arizona and Las Vegas, Nevada

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Registered by TID Oak Ridge, TN as CONF-710809
Library of Congress Card Number: 73-79873

Printed in the United States of America
May, 1973

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INTRODUCTION

This book is prepared from papers presented at the Tritium Symposium which was held on August 28 to September 3, 1971 in the Frontier Hotel, Las Vegas, Nevada. The Symposium was co-sponsored by the University of Nevada, Las Vegas (UNLV) and the Western Environmental Research Laboratory of the U. S. Environmental Protection Agency (EPA). Subsequently, this Laboratory has been renamed the National Environmental Research Center-Las Vegas (NERC-LV).

The organizers of the Symposium hoped to encourage discussion between scientists of various disciplines and, thus, eliminate a number of misconceptions in this field. The organizers also hoped that the "proceedings" of the Symposium would be a helpful document useful to both specialists and beginners in this field.

In preparing this book the Editors have tried to subject the papers to a rigorous review. This process was the main reason for a reduction of the number of pages from an estimated 1700 to the present size without a significant loss of content. A careful observer will find several papers in the book which were not included in the Symposium program. These papers cover areas which, in the judgment of the Editors, were inadequately covered by the program. Unfortunately, the Editors realized very late that deadlines regarding manuscripts would not be met in some cases and, therefore, several presented papers are not included in this publication. The Editors hope the book has not significantly suffered as a result of this compromise.

The Symposium was co-chaired by M. W. Carter (NERC-LV) and R. B. Smith (UNLV). The organization of the scientific program was carried out by the Program Committee composed of the following members:

A. Alan Moghissi, Chairman
Chief, Radiological Research Program
National Environmental Research Center-Las Vegas, EPA
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We are indebted to the members of the Program Committee and to the following individuals who chaired various sessions:

E. A. Evans, L. E. Feinendegen, W. H. Langham,
W. A. Mills, K. Z. Morgan, R. V. Osborne,
H. M. Parker, C. M. Patterson, H. T. Peterson,
C. R. Porter, E. Rapkin, B. W. Shore, R. B. Smith,
and R. C. Thompson

The operation of the Symposium with approximately 400 participants from many countries and the preparation of this book were associated with a number of problems. Credit is due to a number of individuals, notably the following:

J. L. Burk, M. Carpenter, J. S. Coogan, G. S. Douglas,
R. L. Douglas, P. T. Fitzpatrick, M. L. Flanagan,
M. R. Gordon, P. M. Hughes, R. E. Jaquish, J. R. McBride,
P. A. McGill, K. R. McNeil, M. G. Mayes, J. G. Payne, Jr.,
J. E. Regnier, C. J. Rizzardi, C. L. Schnackenberg,
R. B. Sommers, B. K. Spavin, R. E. Stanley, R. D. Tate,
W. J. Wandersee, P. M. Willmore, and P. J. Wunder.

The Editors would like to express their particular appreciation to Raymond T. Moore who, as the Director of the Bureau of Radiological Health and subsequently as the Acting Commissioner of the Environmental Control Administration of the U.S. Public Health Service, supported the Symposium; Stanley M. Greenfield, Assistant Administrator for Research and Monitoring of the EPA, for his enthusiastic support; Patricia Clausen and Jul[illegible] Lashua who were instrumental in the operation of the Symposium and the preparation of the book; and numerous reviewers who helped to reduce the material to a manageable size.

Our thanks to the two most patient people in the world, Barbara Moghissi and Ann Carter.

A. A. Moghissi
M. W. Carter

CHAPTER ONE
Historical and Current Perspectives

HISTORY OF TRITIUM

W. F. Libby
Department of Chemistry
University of California
Los Angeles, CA

It was just forty years ago that I entered graduate school at Berkeley and a good many of my friends here tonight I have known for over twenty years. Much of what I have to say about the history of tritium many of you will remember, in part at least: I think of Wilfred Mann of the Bureau of Standards; Ed Martell, my graduate student at the University of Chicago; Herb Parker from Hanford; Leona Marshall; and many others. And I hope that in these thirty minutes or so of reminiscing that you will note any errors I make for I do think that we should write down before it is too late, the story of the discovery and development of tritium.

I was the first radiochemist at Berkeley. I built the first Geiger counter in this nation. I was in a lucky position to observe development of tritium and so much of what I say tonight is due to happenstance and circumstance. I was there when it happened. I never understood why the true discoverer of tritium, Luis Alvarez, had never written this story. I guess it is because he has had so many great discoveries. I nominated him for the Nobel Prize and the discovery of tritium was number three on the list of accomplishments of that great man. I understand you invited him. I am very sorry he could not be here this evening.

The story begins with one of the last scientific experiments done by Lord Rutherford. He was so prolific, even in his ancient years shortly before his death, he was still producing meaningful nuclear science. The story of tritium begins in 1932 with the development of the artificial accelerators, in particular Ernest Lawrence's cyclotron and the Cockroft-Walton machines. The simultaneous development of the method for making heavy water, deuterium oxide, also happened at that great place we call Berkeley, our alma mater. Gilbert Lewis developed the electrolytic method of separating heavy water, following the discovery of a former student, Harold Urey, that deuterium, the heavy isotope of hydrogen, did indeed exist in nature. Lord Rutherford's experiment was to take the accelerator at Cambridge and to use the deuterons from heavy hydrogen to bombard a target of heavy water. Now Ernest Lawrence had received from Gilbert Lewis the first of the heavy water. . . not the first, the second. The first thing Gilbert did with the heavy water was to feed a good gulp of it to a mouse to see what it would do to him. The second thing he did was to give it to Ernest Lawrence. These are some facts that are not recorded. And Ernest put it in his cyclotron and found it to be a far more prolific producer of radioactive isotopes than ordinary hydrogen, the proton: far more. In that same year, the neutron was discovered. We were not clear at that point of time why the deuteron was so much more prolific as a producer of isotopes. Now we know of course, in our infinite experience and wisdom, that it is the extra neutron in the deuteron nucleus.

But to go back to the Rutherford experiment: he did the seemingly simple experiment of bombarding a deuteron with a deuteron and, with the care which was characteristic of the Cambridge Laboratory, observed that the products of the reaction were such that he had discovered two new, shall we say atoms. He discovered a mass three isotope of hydrogen, one heavier than deuterium, which we call tritium and he had discovered a mass three isotope of helium, one lighter than our ordinary helium. That was about 1934. I have never understood why we never did that experiment at Berkeley. It is very interesting: the timing. Of course everything was extremely hectic in those days.

I will tell you a little story on the side. Ernest Lawrence had the most powerful accelerator in the world, and they were using his deuterium beams to bombard everything they could get their hands on, and were following the Rutherford tradition of observing what came out. And, of course, the physicist graduate students were all standing around with their ion chambers making measurements of what the scattering angles were and so on. Ernest got a cable one night from Joliot Curie. It said, "I irradiated aluminum with polonium alpha particles and made phosphorus-30 (I think it was), which emits positrons with a 2-1/2 minute half-life." This was the first discovery of the production of an artificial radioactivity. Ernest Lawrence went into the laboratory that same evening and discovered radiosodium. The fact of the matter is, the laboratory was so full of artificial radioactivity that it was a health hazard. Malcolm Henderson, who was one of those graduate students, had his white cell count drop to a dangerous level; it even matched mine. I was doing radioactive work with natural radon and we were both ignorant and enthusiastic and not sufficiently careful about the dangers of radiation. It is for that reason I suppose that we did not do the Rutherford experiment which is such a straight forward and obvious one - we were bombarding every element in the periodic table, why didn't we bombard deuterium? Rutherford did it though, and he found that the radiations that came out of the collision were neutrons and protons; we deduce the production of both tritium and helium-3.

Now, I find it interesting, in looking at the careers of great men, to see what mistakes they have made. Rutherford made a mistake. He knew that of these two new isotopes, one must be radioactive and one must be stable. He made the wrong guess. He guessed that it was tritium that was stable. So he started the following titanic experiment. Using his great influence, he got the Norsk Hydro to collaborate. At that time it was the only commercial source of deuterium in the world. It was a Norwegian heavy water plant where they had large electrolytic facilities and could use the Lewis process for enriching the deuterium. They had been making nitric acid for many years with their hydro power, so they rapidly became the source of heavy water the world over. And as most of you know, in World War II this played quite a role in the attempted development of atomic energy in Nazi Germany. But in this early time of 1936, Lord Rutherford got them to take the very large quantity of 99.99% pure heavy water and further electrolize it, looking for tritium. They did, and found that the electrolysis proceeded properly, but they had no method of telling whether there was any tritium in it. So they sent the concentrate to Lord Rutherford who gave it to F. W. Aston, inventor of the mass spectrometer, who searched with his most sensitive techniques. (I also believe there was a physicist at Princeton involved in this experiment). Anyhow, it turned out negative, to two parts in a hundred thousand; the tritium was lost in the deuterium. Going back, they understood enough about the physical chemistry of the electrolysis to be able to calculate the significance of this number in terms of natural abundance and to set a limit.

Now at this time, Luis Alvarez came into the picture. Having heard of Lord Rutherford's work, he realized, with his keen mind, that this great man had made the wrong guess, that the explanation of the failure to find tritium in this very expensive concentrate was that it was tritium that was radioactive and not helium-3. And the measure of the greatness of the man is that he looked around the Lawrence Radiation Lab. I do not think anyone in that laboratory had ever thought of doing this: Alvarez made a mass spectrometer out of the cyclotron. He bent the frequency so it matched the mass helium-3. Then he fed in ordinary helium in the ion stores, put in a sensitive ionization chamber and sure enough, he found helium-3 ions. This broke the back of the problem. He found, in fact, that of ordinary helium in the atmosphere, out of one million atoms, one is helium-3. He found that helium from gas wells is about ten percent as concentrated. But the fact that it occurs in gas wells proves immediately that helium is a stable isotope, or else has a very long life time. The next step was to make tritium and to see if it was radioactive, which he did. He and Robert Cornog manufactured tritium and as I recall, the way they did it was not with bombarding heavy water with deuterons, but with neutrons on lithium. A number of reactions are possible if you look at the isotope table. And he took the gas, having bombarded metallic lithium with neutrons (neutrons produced by the cyclotron), reacted it with water, and used the gaseous hydrogen in an ion chamber and established its radioactivity.

We had no knowledge of the life time except that obviously it must be more than a few minutes or hours, because they waited to see whether the radioactivity decayed, and could observe no decay over a reasonable period. I had been talking to Bob Cornog, Luis' colleague in this, and suggested that we look for natural sources of tritium, for the helium-3 in the atmosphere was ten times as concentrated as the helium-3 in oil well helium. And having studied the isotope table, we wrote down a few possible reactions and discovered in 1939 that both nitrogen and boron, when irradiated with fast neutrons, produced tritium. We did this by irradiating salts and putting the water vapor from these in a Geiger counter, (we simply heated ammonium nitrate and boric acid after irradiation to obtain the water vapor). Strangely enough, it worked. We had a Geiger counter six feet long, six inches in diameter with several millimeters of water vapor; with it we measured the fast neutron production cross sections.

We got very interested in tritium from a chemical point of view and in 1939 and 40, measured the vapor pressure of the liquid hydrogens containing tritium. The hydrogen molecules, you see, are quite numerous: you have H_2; then you have HD where D is deuterium; then you have D_2; then you have HT where T is tritium; then you T_2; then you have DT, and so on. For understanding the nature of the liquid state, an enormously enticing opportunity was to measure the vapor pressures of these different liquids, and we did that. In the course of doing this, we learned how to count tritium better than with our water vapor counter. Luis had used an ionization chamber in his discovery experiment. An ionization chamber, as I am sure many of you in this room know, is not the best way of counting tritium. We developed more sensitive techniques using hydrogen gas in Geiger counters. This served us well in later work with tritium, and it was far more sensitive than the previous methods.

Now after World War II, we decided to go back to the matter of natural tritium. Bob Cornog and I had shown that the cross sections for fast neutron production were appreciable and the same kind of cosmic ray bombardment of the atmosphere, which in the later years was to be shown to produce radiocarbon, should produce tritium.

So we started a search for natural tritium, which was the same thing Lord Rutherford had been trying to do, except that we now knew that it was radioactive. That meant we must be careful not to look at anything except relatively fresh rain water, for if tritium was in nature, it had to be produced by the cosmic rays bombarding the air. You remember perhaps, that I have told you we succeeded in making tritium from nitrogen by fast neutrons. But how to find natural tritium? In the first place, we did not have any idea as to the spectrum of the neutrons produced by the cosmic rays. In other words, we did not have any real idea as to what the production intensity was; we did not have any real idea as to how much tritium we might expect. It was pretty easy to show that the amount must be far below the most sensitive Geiger counter we could invent at that point in time. So we went to our friends in the Cavendish Laboratory who knew about Lord Rutherford's samples (and I would at this point in time thank Dr. A. V. Grosse, Sir John Cockroft, Sir James Chadwick, Dr. Feather, Dr. Thode, Dr. Oliphant, and several others who helped us get our hands on the Rutherford samples). Lord Rutherford had looked at them with a mass spectrometer. We wanted to look at those same samples with a Geiger counter. I recall with a great thrill still, the first time we put hydrogen in our Geiger counter from those Rutherford samples. We got no count. We did not even have a background. It is a characteristic of a Geiger counter when it is overloaded with radiation that it does nothing. So we diluted it with ordinary hydrogen. We got a distinctive response at the counter's maximum rate. The sample was so radioactive we had to dilute it by a very substantial amount, I think it was a thousand fold, in order to be able to measure it. The Rutherford Laboratory was full of instruments that could have measured it. There is a kind of point here that even the greatest men, and the Lord knows there are few men greater than Lord Rutherford was, have a blindness in certain areas. There is the simple experiment of Joliot Curie in which he takes an aluminum foil and puts it on a polonium source which every laboratory in the world had, then puts it on a Geiger counter, and discovers artificial radioactivity. Ernest Lawrence had so much artificial radioactivity he was practically killing his graduate students. Even Fermi did similarly. Fermi thought the fission products were transuranics. We have to remember this every time we look at the history of science.

Well, anyhow, we finally got our hands on the sample and measured it. Now we made Rutherford's original calculation, or I should say Norsk Hydro calculation on fractionation factors on the electrolysis. So from our counts we could go back and roughly calculate the natural abundance at that time. You see, that sample had been made in 1935, we did our work in 1949, and by this time, we were beginning to know what the half-life of tritium was, namely around twelve years. So we had to correct by a factor of two for decay. Thus we go back, and from this we can calculate that the rain water in Norway which was used in the electrolysis plant had about one tritium atom in every 10^{18} hydrogen atoms. That has stood up over the years since as being within a factor of two or three of the correct figure for rain water, undisturbed by human activities, as produced by cosmic rays.

However, we had a queasy feeling about this because by this time we knew that the radiation of heavy water with neutrons would make tritium. And it was by no means clear that the Cockroft-Walton accelerator in Cavendish had not done something to these Norwegian heavy water samples. So we went back to Norsk Hydro and in 1949, persuaded them to make us a duplicate of the Rutherford sample, which they did. And to this day, we are grateful to those gentlemen, the engineers and directors of the Norsk Hydro who took a substantial volume of heavy water, electrolyzed it down, and gave us the concentrate. They started with about three liters of 99.74% D_2O, reduced it to a volume of $15cm^3$ that was one sample. Another sample, they took the electrolytic gases, and gave these.,.,well, leave out the details, the result is that the analysis of the Rutherford sample was correct. We even had the decay correction right. The water which was made fresh for us was twice as hot as the Rutherford sample which had been made fifteen years before. So natural tritium was discovered.

Now all the while we were doing this, there was a German team, under Dr. Paul Harteck, working on hydrogen gas from the atmosphere and as such things go, they discovered tritium simultaneously. In fact, they published a little before our publication, entirely unknown to us, with exactly the same goal except they had a different approach. They took the hydrogen gas from the atmosphere and converted it into ethane, and put it in their Geiger counter and found it radioactive. So tritium occurs in nature.

We then started a massive program to measure the amount and the concentration and to see what we could do with it. We did a study of the electrolytic separation factor so we could measure quantitatively, and convert the count rates into the concentration of the initial feed water. We could do a fairly accurate job of seeing how much tritium was in the concentrate and our friends in the Bureau of Standards helped us by furnishing standards to calibrate our counters. We had to make a calibration however, of the electrolytic cells so we would know when the water volume was reduced say from three liters to fifteen cc, by what factor the tritium would have been reduced. This is an interesting exercise in physical chemistry; I will not go into the details, but in this way we managed to get all the techniques for measuring natural tritium in all natural waters.

Now this all implies a dating technique for water, in that fresh rain is the most radioactive and water that has been bottled loses it with a half-life of twelve and a half years, the tritium half-life. I will never forget my experience of approaching the Air Force which was supporting this research. I went to Washington and said I had to have ten cases of vintage wine from the best thirty-five vineyards in the world because this was the only way I could get rain water of known age, the only way. And they said prove it. So I put an ad in the *Chicago Tribune* and I got three hundred replies; we sorted every one of them through and there was no single place where we had a series of water samples with firmly known age with which we could calibrate our dater. And they were convinced. So the Air Force sent us ten cases of very fine French vintage wine. And we did find that the twenty-year-old wine had about one third the activity of the new wine of the same vineyard. In fact, we could take them from the various years and plot out the decay curve of tritium. Some years later, the Customs Bureau used our method for checking import liquor. I think they may still be doing that. But it was a method of dating wine and rain water up to about fifty years. Also agricultural products. The only agricultural product we have found worth dating so far has been wine.

We have used it very enthusiastically and I hope effectively on studying the atmospheric circulation patterns for water. For example, we found that the rain water in California and on the West Coast of Europe was a lot less, a lot lower in tritium concentration than that in New York or the East Coast. We explained this as being due to re-evaporation. The water which falls on California has been freshly evaporated from the ocean, and has had only one chance to get tritium from the high atmosphere where it is made by the cosmic rays. The New York and Eastern Seaboard rain has had three or four chances, so it runs about three or four times as concentrated in tritium.

This has proved useful in hydrology also. At Lake Michigan, we studied the hydrology and found that the tritium concentration in Lake Michigan was a little less than one-third that of the rain. The rainfall is almost six tritium units and the lake somewhat less than two. Taking the inflow pattern from the drainage area, evaporation, the depth of the lake and the rainfall, one can make a material balance and find that the calculated ratio is almost exactly in agreement. So it looked like natural tritium would be a useful hydrological tool. In the meetings here these last few days, we have had several papers on the use of tritium for hydrology.

We still have not used purposefully injected tritium for hydrological purposes. It is interesting this has not happened. One of the reasons is that the bomb tests have come along and so loaded the rain with tritium that it is a lot easier to measure the tritium in rain than it was in 1953 when we started this research before the bomb tests. And so there has not been much pressure for seeding, but there still is a possibility of getting detailed hydrological information by a purposeful injection of tritium. The sensitivity of the technique we were using was so vast that we needed only very low levels in any case.

The isotope has tremendous powers for medicine and biology. It has one of the softest radiations known. I know of only one other isotope that has softer radiation. It is rhenium-187. This is an advantage for biological purposes. It means that if we can localize the tritium in a particular case, we know where the radiation effect is. I have the greatest hopes for this application of tritium in cancer therapy.

As a tracer in the understanding of the mechanism of biochemical reactions, tritium ranks second only to carbon-14. These are the two we found at Berkeley. We had the inside role there for we had the only mass producer of isotopes in the world. People like John Lawrence, Ernest's brother, were enthusiastic about testing medical uses. All the rest of us had to do was make isotopes, and give them to these doctors; they would explore uses for them. That is the way it all began. We made phosphorus-32 and sulfur-35, but tritium started to rank with these leading isotopes before the final chapter was written. Of course an enormous amount already has been done, using tritium in measuring blood volumes and elucidating the biochemical mechanisms.

Now in the future, we will see if tritium does not prove to be a major source of electrical power. If the controlled thermonuclear reactor is ever developed, we would be going through tritium on the way to the release of fusion energy. So I think this means we should think about what problems might come up when we are using tritium in quantities that make our present activities look miniscule. What problems might come up, I noticed with pleasure Dr. Greenfield mentioned this point in his opening lecture, and there is little doubt in my mind that these problems can be handled, but they should come into the cost estimates that are made at the beginning and not be dragged in later, reluctantly. Tritium is a very benign isotope. Its specific radiation intensity is very low and its radioactive lifetime is short enough so it is something with which we do not permanently contaminate the landscape. But it is very likely to become a major carrier for the production of electric power for mankind in the future. Therefore, we must come to understand how to live with tritium and really understand it. We must not do as poorly as we have done with fossil fuel and forget about the SO_2 until we are dying and choking over it. Our record of behavior in pollution problems is a very poor one. We nearly always have been coerced into it. The only exceptions are the fission power reactors for which we really tried to do something. But fossil fuels, particularly at coal power plants, received minimal planning for solving problems of the pollution of the environment. We must not do that with tritium, though tritium is not so dangerous.

I will close by telling you of another research we did with tritium which applies especially to the origin of the atmosphere and the nature of its behavior. When it decays, tritium makes the isotope, helium-3. And as I said, Dr. Alvarez and Dr. Cornog showed that isotope, helium-3, is tenfold more abundant in atmospheric helium than it is in petroleum or oil well helium. Presumably, the latter is derived of uranium and thorium deposits in the earth. So there must be a source of helium-3 in the atmosphere. It is fairly obvious, since we found tritium in the atmosphere, that there is, in fact, such a source of tritium. From the measure of the amount of tritium in rain water, we can calculate how much tritium is made. Just multiply the content by the annual rainfall and we have the annual production of tritium. Now this lies around in rivers and lakes and decays into helium-3, which then bubbles off and goes into the atmosphere. So we have a source of helium-3 from the tritium production. In addition, it seems most likely that the reactions by high energy cosmic rays which give tritium would also produce helium-3 directly. But in any case, we get a lower limit for the production of helium-3 by just taking the tritium alone. Now compare that with the amount of helium-3 in the atmosphere to get the lifetime of helium-3 in the atmosphere, at least an upper limit to it. Any additional production by the direct processes will only shorten it. Thus we get a figure of ten million years. Now for helium-4, the other isotope, we figure it to be a hundred million, as best as we can tell from the amount of helium in gases escaping the earth's surface, principally the oceans. This figure indicates that the mechanism of gaseous escape from the earth is a thermal one, just as had been postulated, but by no means generally accepted. It must have a very large dependence on the mass of the escaped atom. Now in questioning the origin of the earth's atmosphere, a very basic question arises: How rapidly can hydrogen, which is generated by geothermal processes, escape? As far as we can tell, a large fraction of the primeval water which was here when the earth was formed has been converted into limestone, and a corresponding amount of hydrogen must have escaped. This is a puzzle of very great seriousness in geophysics and geochemistry. The tritium results have led us to rely at least on a hot upper atmosphere, and this led us not to rely on such magic hand waving as solar flares, sudden storms and phenomena with which we are not fairly familiar.

A few years ago, we had a satellite which just happened to be in orbit when a solar flare occurred. It was brought down and examined. This was not our research but we were keenly interested in it. Ed Fireman and Dr. Zahringer, of the Max Planck Institute, independently examined the metal parts of this satellite and showed that it had appreciable concentrations of both tritium and helium-3. The abundance ratio of tritium and helium-3 in our atmosphere is like a million to one relative to helium-4; it was like a thousand to one in the satellite. The concentration of tritium and helium-3 in this piece of metal that had been exposed outside of the earth's magnetic field to the sudden burst of radiation coming from the sun was so hot that we can only conclude that the solar flare, the giant solar storms, probably are manufacturing these two isotopes. In other words, they are doing the Lord Rutherford's experiment of deuterons, or shall we say protons and alpha particles are more likely, but the storms are getting the similar result, producing the same two products. This has led us to understand that these flares are giant accelerators. Let us re-examine the question of whether the main part of our tritium and rain is coming from fast neutrons, ordinary cosmic rays, or whether it is coming from solar flares. Now we are at the edge of our knowledge at this point; I only say for the future that we expect to find, from the moon rock research and from satellite research, more and more information about the past behavior of the sun as an atomic accelerator and to further answer the question of whether natural tritium is produced solely by cosmic rays or if solar radiation is to be involved as well.

I think I should not close without mentioning some of the chemical properties of this wonderful isotope. I remember one time when I was an AEC commissioner, I asked the boys at Oak Ridge to make me a tritium water sample. They did all right. It glowed like an electric light bulb. They had it perpetually in liquid nitrogen in order to keep it cool. Tritium is quite a substance. It is not normally used in that manner. We use it normally in dilute concentrations in order to handle it. But it has been very useful in order to study the effect of mass on hydrogenous molecules. I mentioned the vapor pressure of the liquid hydrogens but similar questions apply to all hydrogenous compounds in their practically infinite variety, experimenting with the deuterium isotope. There is an enormous amount of additional research in this area, studying the intricate details of the thermodynamics and kinetic properties of hydrogenous molecules. The story of tritium is certainly not complete. I think we will all see other things happen. We have in this symposium several recognized important potential applications, to which we look forward with the greatest of interest.

Industrial isotopes, industrial applications of tritium, cannot be entirely neglected in a meeting of this sort. Of course, I have mentioned the generation of fusion power but it is just an intermediate there. I have been a little disappointed that we have not found more really good industrial applications for tritium. We have found a few. Those here who may have some other good ones in mind, I would appreciate very much your telling me about them. It is good in activating flourescent paint replacing radium in wrist watches. It is good in tracing and measuring the flow of fluids, water and hydrogenous fluids, but by and large, I think we need to put our minds more intensely to the question: What can we do with tritium, which is very harmless, very cheap? Why do we not make some real effort to develop additional uses? Certainly in the research lab, it is a wonderful and indispensible tool. And I think it ought to have more applications in industry.

The measurement techniques have been proved to the point now where we can make measurements in numbers and at costs which are an order of magnitude less than twenty years ago. In our exhibit room out here, there are instruments which are examples of what I am saying. In my laboratory we still use Geiger counter techniques together with some of the electrolytic, and they are very laborious. I was talking to the Packard people this afternoon and if they can add on to their instrument catalogue some electrolytic concentrating equipment, we could get down to the levels of tritium which now occur in natural water, which are about twenty tritium units -- about ten times the cosmic ray levels. This rise is due to atmospheric bomb testing. It lasts for many years, but it is falling all the time. Unless we start seeding hydrological applications, we are going to have to go back to the sensitivities we used in the 1951's and 52's. And that is hard work. So I think we do need some commercialization of these laboratory techniques which have been proven to be adequate but very laborious.

It is quite clear we can trace the movement of underground waters. You see, I can take a sample of water and tell to some degree how long it has been underground or else tell something about whether it has recently fallen as rain, using [illegible] as it is, but seeding so greatly adds to the power of the method that this amplification seems inevitable. Now we are going to have to get some information on how to seed some of these water bodies. We worked on Lake Michigan in those bright and early years and wondered whether we should seed Lake Michigan. It is quite a problem: how to mix that mixture? We did research on Crater Lake in Oregon where one of the bomb tests seeded the lake and we watched it mix. It was not all that instantaneous. Now if we have an underground water body, and want to label it to see where it is going, not only do we have to get the permission of the Health authorities, we have to figure out how to stir it. Right at this point in time, we are dependent upon the hydrological applications of what is there now. And it is going to require an order of magnitude of improvement in freshly available commercial instrumentation, which could be done I am certain. If we can do it in the lab, it can be done commercially. These electrolytic cells are not all that difficult to build. I see out there in the exhibits they are taking organic compounds and combusing them to water in one vial and CO_2 in the other vial, and if they can do all that, they can certainly electrolyze water. This will help insure tritium of having a glorious future.

ENVIRONMENTAL PROTECTION AGENCY'S ROLE IN ENVIRONMENTAL TRITIUM

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I am very happy to be here this morning and to welcome you on behalf of the Environmental Protection Agency. This week-long meeting is the first technical session on radiation problems to be co-sponsored by EPA. It will not be the last. I can assure you that this new agency is very interested in environmental radiation. In our zeal to restore the quality of our lakes and streams and to cut down drastically on industrial and automotive air pollution, we will not overlook or downgrade the hazards of radiation.

Indeed, the organization of EPA as an agency separate from the long-established government departments and reporting directly to the President gives us a clear mandate and a clear opportunity to act independently of those components of government and sectors of our socio-economic system which are likely to have a vested interest in promoting some polluting activity.

As Bill Ruckelshaus put it: "The only thing EPA is promoting is a better environment."

I think we have a tendency to underestimate the hazard of tritium for several reasons:

First, we are making so much of it. Nuclear power plants produce tritium both from the fissioning of the uranium fuel and from neutron capture reactions in the coolant and in the core materials. Tritiated water constitutes almost all of the radioactivity in liquid waste and most of the radioactivity in gaseous waste discharged to the environment from a typical nuclear power reactor. A single power plant will discharge hundreds of curies per month in tritium, tenths of a curie per month in all other radioisotopes.

Our early experience with nuclear fuel reprocessing plants indicates that these plants will be even more prolific producers of tritium.

Second, the principal form of tritium in reactors is tritiated water. Conventional concentration processes, such as ion exchange and evaporation, are not effective at reducing its volume for permanent containment. It is released as an inseparable part of a basic constituent of the environment.

Third, because it is in all water, tritium is ubiquitous. It is everywhere in living matter. We cannot shield ourselves from it; we have to live with it, and by it.

Fourth, problems associated with detection and quantitative measurement.

The Atomic Energy Commission sets stringent rules for each power plant's radioactive effluents, including tritium. But the tritium standards allow discharges that are several orders of magnitude greater than other radioactive wastes. Until recently the liquid wastes from nuclear reactors were not routinely monitored for tritium; the discharges were calculated from operating data. They have always been comfortingly low when expressed in a percentage of the discharge limit.

I believe it is time we paid more attention to tritium. It is not yet an acute problem from the environmental standpoint, but it can become one.

Our information on how much tritium occurs naturally in the biosphere is partly conjectural, since we did not start to measure it carefully until after the contamination from nuclear weapons began. The best estimates fall in the range from 16 to 35 picocuries per liter of water, or, in more absolute terms, 5 to 10 tritium atoms for every 10^{18} atoms of normal hydrogen.

The natural production of tritium - from cosmic radiation of oxygen and nitrogen in the air - is believed to be constant and to account for about 69 megacuries in the biosphere at any one time. Weapons testing produced 24 times as much before the atmospheric test ban treaty went into effect in 1962!

Since then, there have been fewer atmospheric tests. But underground detonations have continued to produce comparable amounts of tritium in the earth's crust. How safely it will be contained there we simply don't know. Some of the underground tests release tritium estimated by the Lawrence Livermore Lab as high as one megacurie per event.

Most of the tritium from nuclear tests was injected into the stratosphere, where we believe it stays for a period of years because of the slow mixing rate with the troposphere. In the lower atmosphere, however, tritium is rapidly precipitated and mixed with surface waters. Rainfall following periods of nuclear testing was found to contain several hundred times the normal tritium concentrations, and the "half-life" in the lower atmosphere seems to be 35 to 40 days.

This shows very clearly how tritium always winds up as water and pervades the environment (with some delay in the stratosphere in the case of nuclear explosion products). Tritium spreads as water spreads. Nature doesn't let it accumulate anywhere. Man has tried to contain it underground with doubtful success, in the case of weapons testing. Tritium has been released in gas stimulation experiments through the deliberate flaring of gas. In the routine production of power from nuclear reactors, no containment is even tried.

Estimates vary as to how much tritium we are creating in nuclear reactors and releasing eventually as tritiated water. It is probably approaching a million curies per year.

There is general agreement that the nuclear power industry is expanding. What will the tritium concentrations in the environment be in 1980? In 1990? What will be the environmental impact of all that tritium? These are questions we need to ask now, while there is still time to exert influence effectively on our nation's environmental planning.

It seems certain that we cannot go on indefinitely with our present method of tritium disposal, that is: loosing it to the environment and hoping it will spread and dilute to levels that are harmless by any operating definition. In the absence of conclusive evidence we assume that there is no truly harmless radiation level, no threshold.

This conference was organized to bring such questions to the fore. Primary among them is the development of the nuclear industry which is producing the tritium. How much will the industry expand?

What changes in reactor operation or design can reduce radioactive wastes?

EPA wants to support and encourage research in these matters with all the effort we can muster.

We want to know more about how tritium reacts in living matter. Is there any biological magnification of radioisotopes along a food chain, similar to the increasing concentrations found with certain chemicals? To put it more precisely, is there an isotopic effect of tritium in living tissue? Most hydrogen in living organisms is in water molecules, but some is in organic and cellular structures, as carbohydrates, fats, lipids, and so on. The proteins of the cell nucleus contain hydrogen. It seems quite possible that at the internal cellular level there may be reactions that discriminate between tritium and normal hydrogen.

Such research would be exceedingly valuable from the standpoint of basic knowledge, but it also could reveal clues to possible methods of tritium control. We are particularly interested in knowing the relationship between aqueous and tissue-bound tritium over several generations and under conditions that are ecologically realistic.

Also, what are the biological effects - the radiation damage - of tritium at low levels of contentration? Research in this area is particularly pertinent because it seems likely that any tritium exposures that develop will be at low levels, to large populations, from widespread environmental sources. The chance of catastrophic accidents to individuals or small groups is very slight with tritium. We are glad this is so, but it means we must start our biological effects research from the bottom of the dose ladder and work up.

Finally, we in EPA want to encourage tritium disposal research *before*, rather than after, it becomes an acute problem. This is a unique opportunity for environmentalists. The AEC and the nuclear industry point out - with some justifiable pride - that no other large enterprise is as careful as they are in studying and perfecting ways to minimize and control their pollutants before they produce them.

This is true in most instances. The nuclear scientists have been keenly aware of radiation dangers and have worked very hard to forestall them.

Tritium is a paradox right now; it is the most widespread man-made radioisotope and the least harmful. Now is the time to find out how to control it.

* * *

THE SOCIOECONOMIC IMPACT OF LOW-LEVEL TRITIUM RELEASES TO THE ENVIRONMENT

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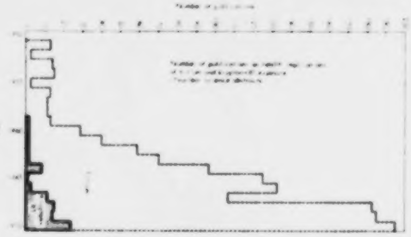
The development of nuclear energy has been accompanied by an increasing concern over tritium releases to the environment. Over the past 10 years research on the subject has proliferated. The rate of publications on health implications of environmental tritium is shown in Figure 1. This information indicates an increase of approximately 20-fold in the number of these publications over the past

10 years. Also shown, for purposes of comparison, is the number of publications related to krypton-85.

Elaborate systems are being developed for the reduction of tritium produced and released from reactors. In the Plowshare Program, large sums of money have been expended toward the development of a low tritium-producing nuclear explosive for gas well stimulation. With all this activity and concern, one might well reflect on the question, "How serious is the tritium problem?" Research begets more research, and quite often we can get so involved exploring the details and intricacies of a subject that we can easily lose our general perspective. It is the purpose of this paper to explore the position of tritium in the scheme of things, or stated differently, to determine the social and economic impact associated with environmental tritium.

Figure 1

Number of Publications on Health Implications of Tritium and Krypton-85 Exposure (Nuclear Science abstracts)



This determination requires some basis for comparison that will be commonly understood; a scale, if you will, on which to measure the seriousness of the problem. For this purpose, we shall make use of a form of benefit-risk analysis.

As has been stated many times, the risk of radiation exposure should be balanced against any benefit associated with such exposure. Some studies involving benefit-risk analysis have already been performed, [1-3] although these are still at a preliminary level. The objective of benefit-risk analysis is to determine rational, definitive, and generally acceptable means of evaluating the potential benefits of any given operation, program, or technology against the possible risks.

Two years ago, the use of a new unit called the "mer" was proposed [4]. The mer is defined as that amount of benefit required to justify an exposure to 1 rem. The tangible and, hopefully, comprehensive mer equivalents proposed were 25 days of life, 2000 motor vehicle miles and, most important, $250. Use of the latter figure would conveniently enable one to calculate, in dollars, the biological cost of various radiation exposures, and compare this to the cost of remedial measures necessary to reduce or avoid such exposures. This approach implies a monetary value for a statistical human life, specifically $250,000 [4].

Some scientists have objected to assigning such values for low-level radiation doses and/or statistical human life [5]. These objections range from morality implications to dispute over linear extrapolation of radiation damage from high to low doses. We can understand most of these arguments, particularly when applied from the viewpoint of any given individual who compares his own risk to his own benefit, using his own prejudice or opinion. If it is averaged over large enough groups, however, we believe this approach is neither immoral nor unreasonable; it is merely the explicit reflection of a type of value judgment which frequently is made by society.

The value of the mer was originally determined partially on the basis of a poll of individuals professionally knowledgeable on radiobiological effects. Subsequently, other authors [6-9], Table 1, using various assumptions, have also provided estimates on the monetary value of biological damage due to radiation exposure. Despite the varying assumptions on which they are based, these estimates show a remarkable consistency. The $250 we have chosen is the highest, or most conservative, of these values. As we apply it, we assume a linear relationship for radiation damage which also results in a very conservative estimate of the environmental cost.

Table 1

Assumed Biological Cost of Radiation Damage

Author	Cost ($/man-rad)
Cohen (LLL), 1969	250.00
Hedgran, Lindell (Sweden), 1970	100.00
Dunster (UK), 1970	~10.00*
Otway (LASL), 1971	200.00
Lederberg (Stanford), 1971	100.00

*A few pounds sterling.

Other assumptions we shall use in our analysis are:

1. All activities are conducted in such a manner that the limits set by all appropriate standards and guidelines are not exceeded.
2. MPC limits are valid indicators of human exposure. Anyone continually exposed to the 168 h/week exposure limit for uncontrolled areas would receive an exposure of 500 mrem/a, and this relationship may be linearly scaled.
3. Any quantity of released tritium could, in time, expose everyone in the world to its potential effect.
4. The environmental half-life for tritium is equal to its radiological half-life, and its mean environmental residence time (MERT) is equal to its mean radiological life. This conservatively infers that no "sink" which takes up tritium in such a manner to make it biologically unavailable exists in the biosphere. For tritium the MERT equals approximately 17 years.

5. Some physical constants required for this analysis are [10-11]
   a. Mass of the atmosphere[10] = 5×10^{21} g
   b. Volume of the atmosphere (at NTP conditions, sea level, 0°C) = 4.2×10^{24} cm^3
   c. Volume of the hydrosphere[10] = 1.41×10^{24} cm^3
   d. World population[11] = 3.5×10^9.

Now, with this background, given the information that tritium has been released to the environment, we can attempt to determine, in dollars, the worldwide biological damage that will be done.

The cost of radiation damage to human population is equal to the product of: Average individual exposure, times the number of individuals exposed, times the number of \$/man-rem, times the average duration of the exposure. The average exposure is equal to the average concentration of the radionuclide in the biosphere, divided by the appropriate MPC, and times 0.50 rem/MPC. For example, the radiation damage cost which can be attributed to the releases of tritium to the atmosphere is:

$$\frac{0.5 \text{ rem/MPC-a}}{4.2 \times 10^{18} \text{ m}^3 \times 2 \times 10^{-7} \text{ Ci/m}^3\text{/MPC}} \times 3.5 \times 10^9 \text{ people} \times 17.0 \text{ a} \times \$250\text{/man-rem} = \$9.0\text{/Ci}$$

Note that the MPC used is that for air. If the tritium were released to the hydrosphere both the volume for dilution and MPC would be different and the value obtained is then only 0.002 \$/Ci.

Tritium released to the biosphere partitions between the hydrosphere and the atmosphere. Jacobs [12] Table 2, predicted dose rates due to accumulation of tritium in the world. He postulated that tritium would accumulate in the hydrosphere, but assumed only the top 75 m of the ocean was available for dilution. He concluded that the annual dose rate from each curie of environmental tritium would be about 10^{-14} rem/a. Using this value in the first term labeled "exposure" in the equation above, we obtain:

$$10^{-14} \text{ rem/Ci-a} \times 3.5 \times 10^9 \text{ people} \times 17 \text{ a} \times \$250\text{/man-rem} = \$0.15\text{/Ci}$$

Table 2

Predicted Effect of Tritium Accumulation in the Biosphere*

Year	Total accumulated Tritium (MCi)	Average dose rate (rem/a)	rem/Ci-a
1970	0.3	2.8×10^{-9}	10^{-14}
1975	2.0	1.5×10^{-8}	
1980	6.0	5.5×10^{-8}	
1985	16.0	1.5×10^{-7}	
1990	32.0	2.8×10^{-7}	
1995	58.0	5.0×10^{-7}	
2000	96.0	8.2×10^{-7}	

*Data from Reference 12.

The present inventory of the world supply of tritium is due to that naturally formed plus that which is man-made, largely from the atmospheric weapons tests of the late 1950's and early 1960's. Begemann [13] estimated this is distributed 99.8% in the hydrosphere and 0.2% in the atmosphere. If new releases of tritium distribute in this way, the average biological damage cost is about \$0.02/Ci.

Tritium formed naturally in the upper atmosphere is distributed approximately 90% in the hydrosphere and 10% in the atmosphere [13]. Assuming this latter distribution, a biological damage cost of approximately \$0.90/Ci can be determined. This information is summarized in Table 3.

Table 3

Environmental Tritium Partition and Effect

Basis	Relative Mix % atmosphere/hydrosphere	Estimated cost of biological damage (\$/Ci)
All Atmosphere	100/0	9.0
All Hydrosphere	0/100	0.002
Total World Tritium	0.2/99.8	0.02
Naturally Formed Tritium	10/90	0.9
Only top 75 m of hydrosphere are available for dilution (Jacobs' assumption)	0/100	0.15
Cohen - Higgins estimated cost	--	0.10

There seems to be no way to be certain which distribution will apply; however, prejudice seems to be that new releases will distribute more like the previous releases of man-made tritium. In any event, the value \$0.90/Ci probably represents an upper limit and the value \$0.10/Ci seems a reasonable order-of-magnitude estimate which we will use in future discussions. This is slightly smaller than the value based on Jacobs' estimate. We believe it is reasonable to assume that over a 17-year period some exchange should occur across the oceanic thermocline. This would increase the available hydrosphere from the 75-m depth used by Jacobs and therefore increase the dilution.

As a comparison, one may apply the same type of analysis to krypton-85 releases. In this case the calculations are less equivocal, since one may safely assume the atmosphere is the only region of dilution. The calculation follows:

$$\frac{0.5\ \frac{\text{rem}}{\text{MPC-a}} \times 3.5 \times 10^{9}\ \text{people} \times \$250/\text{man-rem} \times 15.5\ \text{a}}{4.3 \times 10^{18}\ \text{m}^{3} \times 3 \times 10^{-7}\ \text{Ci/m}^{3}\text{-MPC}} = \$5/\text{Ci}$$

One practical application would be a quantitative evaluation of the radiation hazard associated with natural gas supplies stimulated by nuclear explosives as compared to the benefit which might be derived. To perform this analysis we can use the model of Werth, et al.[14] who considered a 28-well, nuclear-stimulated (N.S.) gas field supplying part of the total supply to the Los Angeles Basin. According to this model, Table 4, over the first six years of operation 3.5 X 10^{11} ft^3(10^{10} m^3) of N.S. gas will have been supplied. This gas will have had an average concentration of 4.25 pCi/cm^3 of tritium and 27.5 pCi/cm^3 of ^{85}Kr. Over the first 20 years of operation, the field will have produced about 9.3 X 10^{11} ft^3 (2.6 X 10^{10} m^3) which may be assumed to be about half the total producible. During the first six years of production, however, the wells will be purged of essentially all residual gaseous radioactivity. Over this period, the biological damage cost due to radiation exposure can be determined for both the local and worldwide population.

Table 4

Nuclear explosive stimulated natural gas supply to Los Angeles Basin*

Time period (a)	Volume supplied (10^9 ft^3)	Av concentration pCi/cm^3 tritium	^{85}Kr	Total curies tritium	^{85}Kr
0 - 6	350	4.25	27.5	4.2 X 10^4	2.7 X 10^5
6 - 20	580	negligible	negligible	----	----
20 - ∞	~930	negligible	negligible	----	----
Total	1860	----	----	4.2 X 10^4	2.7 X 10^5

*Based on model of Werth, et al., for 28-well gas field. (~9% of total L. A. supply for initial six-year period.)

Neiburger [15] estimates that under average Los Angeles meteorological conditions, the daily dispersive air volume ≃1.6 X 10^{12} m^3. Based on an average of one air change per day, over the initial six-year period this would amount to a total of ~3.5 X 10^{15} m^3. Therefore, for tritium the cost of biological damage to the local population (risk) would be:

$$\frac{4.2 \times 10^4 \text{ Ci} \times 0.5 \frac{\text{rem}}{\text{MPC-a}} \times 7 \times 10^6 \text{ people} \times \$250/\text{man-rem} \times 6 \text{ a}}{3.5 \times 10^{15} \text{ m}^3 \times 2.7 \times 10^{-7} \text{ Ci/m}^3\text{-MPC}} = \$2.3 \times 10^5$$

The above calculation assumes that all the tritium is released to the air only and, for local population, all exposure is via the air. For ^{85}Kr these same conditions would obviously hold, and the cost (risk) would be:

$$\frac{2.7 \times 10^5 \text{ Ci} \times 0.5 \frac{\text{rem}}{\text{MPC-a}} \times 7 \times 10^6 \text{ people} \times \$250/\text{man-rem} \times 6 \text{ a}}{3.5 \times 10^{15} \text{ m}^3 \times 3 \times 10^{-7} \text{ Ci/m}^3\text{-MPC}} = \$1.3 \times 10^6$$

It is realized that exposures to the local population would not be uniform, and that certain individuals might receive doses far in excess of the average while others would receive far less. As previously stated, however, it is assumed that all exposures would remain below MPC levels. In fact, Werth, et al., [14] estimate that under conditions assumed in the model, the maximum individual exposure (unvented heaters, tight buildings, etc.) would be 2.4 mrem/a, well within exposure guidelines.

For worldwide population exposure, the biological damage costs would be:

For tritium: 4.2×10^4 Ci X \$0.10/Ci = $\$4.2 \times 10^3$

For ^{85}Kr: 2.7×10^5 Ci X \$5/Ci = $\$1.4 \times 10^6$

A summary of these costs is given in Table 5 which also shows the benefit from supplying this gas. This benefit is exactly equal

Table 5

Summary of data from Los Angeles Basin 28-well N.S. gas field (Werth Model)

A. Biological damage costs (risk), in dollars

	To Local Population	To Worldwide Population	Total
Tritium	\$ 230,000	\$ 4,200	\$ 234,200
Krypton-85	\$1,300,000	\$1,400,000	\$2,700,000
Total	\$1,530,000	\$1,404,200	\$2,934,200

B. Benefit (assume price of gas to consumer = \$0.60/MCF)

Period	Total gas supplied	Consumer cost (benefit)
Initial 6 year	3.5×10^{11} ft^3	$\$2.1 \times 10^8$
Total	1.86×10^{12} ft^3	$\$1.1 \times 10^9$

C. Benefit/risk ratio

1. Only initial 6 years considered

$$\frac{2.1 \times 10^8}{2.9 \times 10^6} = 72.4$$

2. Over total production period of gas field

$$\frac{1.1 \times 10^9}{2.9 \times 10^6} = 379.3$$

to the price paid for it. If it were less the customer would not buy it; if it were more the suppliers would demand a higher price. (This is exactly true only in a free market. Natural gas prices are regulated, and for this commodity the value is only approximately equal to the price.) Thus, for an average consumer price of gas of \$0.60 per 1000 ft^3 or more and an increment of gas equal to 350 billion ft^3, the benefit is \$210 million. The benefit-risk ratios are therefore 380 or 72.4, depending on whether the entire production period is considered or only the six years during which radioactivity is released. The ratio indicates that producing incremental gas via nuclear explosive stimulation may easily be considered of overall societal benefit, even though it carries some radioactivity. This conclusion would still be justified with quite different and larger values for the mer.

A "best environmental choice" of energy sources for a given consumer area requires benefit-risk analysis for each source. It is beyond the scope of this paper to perform a complete benefit-risk analysis of nuclear gas stimulation, but one other energy source can be compared using this approach. Nuclear power reactors produce almost 10^{-3} Ci of tritium and 0.02 Ci of ^{85}Kr per MWhe. According to our previous example, a Plowshare-associated natural gas would contain about 10^{-4} Ci of tritium and 0.008 Ci of ^{85}Kr per MWh (1000 $ft^3 \simeq 10^6$ BTU $\approx$ 0.3 MWh). It can therefore be seen that per unit energy produced, the gas would be a better environmental choice. We must hasten to add that this is not a condemnation of nuclear power reactors. The electrical energy from reactors is, of course, not the same as the thermal energy from the combustion of gas. At current prices, per unit of energy, the electricity sells for about five times more than the gas. Additionally, not all the tritium and ^{85}Kr is released from a reactor, although practice varies from reactor to reactor and time to time.

Currently, and for the foreseeable future, the major producer of radioactivity will be nuclear-fission-powered reactors. Table 6 shows the quantities of tritium and ^{85}Kr produced in typical reactor operations [16]. The relative biological damage cost due to release of these nuclides is \$2.50 and \$2500 per MWe-a, respectively. These figures, when viewed in light of those in Figure 1, indicate that perhaps our priorities have been misplaced. The effort devoted to the study of the biological effects of tritium seems out of proportion to those devoted to ^{85}Kr, and the expenditures made to reduce both may be out of all proportion to their impact on mankind. This would be especially true if suggestions for the elevation of the MPC values for tritium [17] by a factor of 10 to 100 and of ^{85}Kr [18] by a factor

Table 6

Quantities of Tritium and ^{85}Kr Typically Produced by Nuclear Power Reactors (Fission)

Nuclide	Production rate (Ci/MWe-a)	\$/Ci	\$/MWe-a
Tritium	~25	0.10	2.50
Krypton-85	~475	5.0	2500.00

of 40 are accepted. Any increase in the MPC will reduce the cost of biological damage per curie.

Determination of the biological cost of released radioactivity in monetary terms can be a powerful tool to use in benefit-risk evaluations such as those now required by the National Environmental Policy Act. At least, it can provide a definitive basis for judgment.

ACKNOWLEDGMENT

This report is based on work performed under the auspices of the U.S. Atomic Energy Commission.

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MAN-MADE TRITIUM

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This presentation is not intended to be an exhaustive treatment of the subject of man-made tritium. Rather, I propose to review in general the major sources and mechanisms for production of man-made tritium. I have also chosen a course of presentation that characterizes the generation of tritium in terms of broad ranges.

The topic of man-made tritium to be discussed divides itself into several subtopics. These include the mechanisms for the production of man-made tritium, the extent of this production as compared to natural means of production, predictions of the rate of tritium generation by human activities in years to come, some problems concerning the management of man-made tritium, and some conclusions as to measures for reduction of tritium in the environment.

Production Mechanisms and Sources of Man-Made Tritium

The principal sources of man-made tritium are electrical power generating reactors, reactors producing nuclear materials for defense purposes, atmospheric tests of nuclear weapons, heavy water plants, nuclear reactors designed specifically to produce tritium, tritium separations plants, and tritium handling operations. In the future, the predominant source of man-made tritium is likely to be generation of electrical power through thermonuclear fusion machines.

Generally speaking, any process involving generation or processing of tritium is a potential source for the release of tritium to the environment. The retention factor for the tritium may approach 100%, but there always seems to be some losses. Processes involving massive amounts of tritium must be especially effective in its retention.

The mechanisms for the production of tritium in nuclear reactors have been discussed by Jacobs [1], and they have been reviewed by Kouts and Long [2]. Briefly, tritium is produced as a ternary fission product in fuel elements of reactors; it is produced through neutron reactions with boron or lithium present for reactivity control, pH control, or other reasons; and it is produced through neutron reactions on other light nuclei such as helium or deuterium. In heavy water-moderated reactors, neutron capture in deuterium generates tritium at a rate well above ternary fission. Jacobs [1] has projected through the year 2000, the worldwide rate of tritium production for all anticipated nuclear power reactors. Although his data show the rate of production, they do not show the rate at which dispersal takes place into the environment. His data reveal the following:

Production of Fission-Product Tritium
In Free-World Nuclear Power Economy

Year	Power Production		Tritium production rate, mCi/a
	GWe*	GWth**	
1965	1.9	6.2	0.03
1970	9.6	31.0	0.1
1975	46.0	150.0	0.7†
1980	120.0	390.0	2.0
1985	240.0***	780.0	3.0
1990	430.0***	1300.0	6.0
1995	670.0***	2200.0	10.0
2000	1030.0	3320.0	15.0
Steady state	1030.0	3320.0	15.0

*Assumed load factor of 0.80 to 1980 and 0.70 at 2000

**Assumed thermal efficiency of 0.31

***Interpolated values

†This is also the approximate total amount currently released to the offsite environment from AEC-owned and operated facilities in the R&D and weapons programs. This number will drop in coming years as additional controls are implemented.

A number of authors have made estimates of the amounts of tritium left in the environment after atmospheric testing of thermonuclear weapons [1]. The residual tritium is estimated by various authors and in various cases to range from about 0.7 kg of tritium to 5 kg of tritium per megaton-equivalent explosion. The observed rate of deposition after the Castle series of tests in 1954 indicated about 1.16 kg of tritium per megaton equivalent. The tritium remaining from earlier atmospheric tests by the U.S., U.K., and U.S.S.R. is now dispersed in the stratosphere, and diffusion into the troposphere is slow. A summary of the status of this residual tritium in the atmosphere has been given by Peterson et al. [3]. It should be noted that the Peterson data represent all tritium from testing, and not just the part delivered to the stratosphere. The latter is approximately half the total.

It should also be noted that Peterson's data show only the effects of atmospheric tests before 1963. The tests by France and the People's Republic of China in subsequent years have not been included.

In commenting on the sources of man-made tritium other than those related to electrical power generation, the Atomic Energy

Commission has a responsibility under law and government directive for ensuring that the nation's defense needs are met. This responsibility requires production of tritium for weapons use and other applications. Under the limited test-ban treaty, the injection of tritium into the atmosphere through U.S. atmospheric testing ended a number of years ago. The activities related to nuclear weapons fabrication for the national defense are a source of some release of tritium to the atmosphere. Tritium is generated in operation of the production reactors at Savannah River, where tritium is also isotopically separated from other hydrogen and is processed in tritium handling plants. These activities of production and fabrication account today for most of the tritium released to the environment by the AEC-owned and operated facilities in 1970 (excepting underground nuclear tests) was considerably less than 1 MCi, i.e., less than 100 grams. The exposure to tritium from cosmic rays and residual tritium from past atmospheric testing results in doses which are minor when compared with those from other naturally occurring sources of radiation to which man is exposed.

There are other minor uses of tritium, such as experimental operations and isotopic applications. For instance, Catlin and Western in 1966 [4] indicated that 83 kCi of tritium were used in products considered to involve so little public exposure to radiation that they are either exempted from regulatory control or regulated with minimal requirements.

Future Sources of Man-Made Tritium

It is interesting to speculate on the problems of tritium generation that will accompany controlled thermonuclear fusion reactors, if and when they become a reality. The fusion reactor most likely to achieve success at this time would use tritium in reactions of deuterium with tritium. The deuterium can be extracted from ordinary water, but the tritium would have to be created by a breeding process based on irradiating ^{6}Li with the neutrons from the basic fusion reaction. A fusion reactor of reasonable size would generate about 3 GWe and would consume about 1 kg of tritium daily. It would, therefore, have to generate at least 1 kg (10 MCi) of tritium daily and this daily production rate from 1 thermonuclear reactor would be four times the annual production rate of tritium in all civil power reactors expected to be in operation in the year 1980. In other terms, the tritium production rate to accompany fusion reactor operation will be about 5×10^4 times that of a fission reactor of corresponding electrical capacity. The containment of this tritium is the principal environmental problem facing the successful development of the fusion reactor.

It has been estimated that by 1975 the U.S. contribution to the inventory of tritium in the environment will be about equally divided between nuclear power generation and AEC activities. This estimate neglects the tendency toward designs of reactors and fuel reprocessing plants favoring near zero release of activity and the similar reduction of releases from AEC activities.

The meanings and relationship of these estimates can be assessed using the data published by Peterson et al., showing estimates of atmospheric inventory from all sources. The inventory from AEC activities and from power reactor operation in 1975 will each be from one to two orders of magnitude below the inventory from cosmic ray effects. They will be less than the atmospheric inventory of residual tritium from atmospheric weapons testing in the early 1960's.

Some Suggestions for Lowering Tritium in the Environment

In conclusion, it occurs to me that there are several major ways to reduce the amount of tritium released in the environment as far as man-made effluents are concerned. Since AEC operations are currently making a contribution of tritium to the environment, the AEC feels it is important to continue its policy of reviewing and improving processes and operational methods to reduce, to the lowest practical levels, releases of tritium to the environment from its operations. For example, from 1968 to 1971, AEC-owned facilities were able to effect a 30% reduction in tritium releases by revision of operating procedures and administrative controls, and by implementing new processing and effluent control systems. The nuclear industry should continue the development of systems for reactors and fuel reprocessing facilities which favor increased control of tritium releases to the environment.

As one can see from the residuals of tritium from thermonuclear detonations, which do not include recently conducted tests by the French and Chinese, a reduction of great significance in the atmospheric inventory of residual tritium could be achieved by conducting all nuclear weapons tests underground.

In connection with expected generation of large quantities of tritium in thermonuclear reactors, there will be considerable incentive, as the value of tritium as a fuel makes its recovery economically attractive, to let as little as possible escape.

I cannot leave you without a word on safeguards. Careful control of man-made tritium in the areas of its generation, storage, and release to the environment will become appreciably more necessary as the strategic value of this material fully emerges, and as the amounts of man-made tritium increase.

Tritium management should be considered in the broad sense of the word, including measurements, containment, report of transfers, surveillance and transportation. You all recognize these terms, used also in connection with nuclear materials safeguards.

There is now defined in AEC regulations, published for public comment, a proposed reporting requirement for transfers and a definition of de minimis quantities not subject to reporting either for transfer or theft. These regulations would apply to licensees. AEC tritium is under similar but slightly more rigorous requirements already.

We are presently developing the pros and cons of a safeguard system for tritium which would be effective, manageable and within or close to the state of the art of measurement and containment, and be feasible for implementation considering the numerous possible sources of generation of such material.

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CERTAIN SUBJECTS OF GENERAL INTEREST ON TRITIUM HALF-LIFE AND NOMENCLATURE

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During the symposium, much of which is the basis of this book, several discussions were held which are of significance to the solution of certain tritium problems. The first subject of interest was the adoption of an "accepted" half-life for tritium. There was a general agreement that the most commonly applied value of 12.262 years [1], is inadequate. Although the value reported by Eichelberger, et al. [2], has never found general acceptance, recently this value seems to have been confirmed by Jordan, et al. [3], who report a value of 12.346±0.002 years. It is of interest to note that earlier results by Jones (12.41 years) and Jenks, et al. [4-5], (12.46 years) are in better agreement with the latest values. Although during the symposium no agreement was reached regarding the proper value, there was a general agreement that a value longer than 12.262 years is more appropriate. A value of 12.30 years or, even better, 12.35 years was considered.

Another subject of general interest was the units used to describe tritium concentration. All methods for tritium measurement, on a routine basis, are based on the radioactive decay of tritium. Therefore, any other concentration unit is calculated from radioactivity measurements. As there is a general agreement on the unit of radioactivity (curie), all tritium results are measured in Ci. In certain branches of science, notably geochemistry, it has become customary to use Tritium Unit (TU) or Tritium Ratio (TR). This unit was originally proposed by Libby and represents the approximate level of tritium in the uncontaminated surface waters. The value assigned to it is a ratio of 1 tritium atom in a mixture of 10^{18} atoms of all the hydrogen isotopes. The major problem with this unit is its inherent inaccuracy due to the required knowledge of tritium half-life. The newest value of $T_{1/2}$ of 12.35 years deviates from the old value by approximately 0.7%. In certain cases authors do not indicate the factor they used to convert the measured radioactivity to TU or TR. For geochemical purposes, however, inaccuracies of 0.7% are well within the limit of acceptance and, therefore, the application of TU seems to be justified.

Another consideration was the notation of this unit. Many participants were dissatisfied with the proposed TR instead of old TU. To avoid the problem, a new name, Libby Unit, was proposed in honor of W. F. Libby, who originally proposed the unit and contributed decisively to the success of tritium research. The abbreviation of Libby Unit would be either LU or the existing TU. In any case, to convert measured values into the unit notation (LU or TU), the following multiplication factors may be applied. If a $T_{1/2}$ of 12.30 years is used, 1 TU corresponds to 3.231 pCi/l of water or

29.08 nCi/g H. For a $T_{1/2}$ of 12.35 years, the values would be 3.218 pCi/l and 28.96 nCi/g H. However, it certainly would be easier if tritium concentrations were expressed in appropriate subdivisions of Ci per unit volume or weight.

A third subject discussed during the symposium dealt with the notation of hydrogen isotopes. Letters T and D have been adopted for hydrogen isotopes 3 and 2. Based on the general guidance of the International Union of Pure and Applied Chemistry (IUPAC) and the corresponding Union of Physics (IUPAP), a letter describing a chemical element represents all isotopes of that element. Therefore, H should be used if one deals with nonisotopic character of hydrogen or if hydrogen isotopes (protium, deuterium and tritium) in a mixture are considered. No letter is known to describe hydrogen-1 unless the general guide is applied and ^1H is used. This latter notation disagrees somewhat with the application of D and T and, thus, one would be tempted to use ^2H and ^3H for deuterium and tritium. However, descriptions such as HTO and D_2O are too common to be eliminated and T and D are more convenient to use, in particular if one deals with labeled compounds. The alternative solution would be to select a letter, other than H, for protium. The application of p, as proposed by Libby, has met with great resistance and contradicts the IUPAC rule which requires the application of capital letters for chemical elements. The letters P, Pr, Pm, Pu and Pt have already been assigned and, thus, cannot be used for protium. In this book the reader shall find inconsistencies regarding this subject as the authors' choices were accepted.

Hopefully, future symposia will find solutions to these problems.

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CHAPTER TWO
Tritium Production

YIELDS OF TRITIUM DURING THE FISSIONING OF NUCLIDES OF INTEREST IN THE OPERATION OF NUCLEAR POWER REACTORS

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The increased demand for electrical power in the world will lead to requirements of greater than 2,000 GWe (GWe = 10^9 watts electric) from nuclear power sources by the year 2000. This demand will bring with it many associated problems. One serious problem is how to prevent an undue increase in the world radioactive background. Tritium is formed in nuclear power sources in both thermonuclear fusion and nuclear fission. Because of its intermediate half-life (12.24 years) and biological importance (an isotope of hydrogen), tritium poses a very special hazard to the world population.

At present it is estimated [1] that the world inventory of tritium probably does not exceed 20 kg. At the peak of atmospheric H-bomb testing the inventory was about 200 kg of tritium. First it will be necessary to decide upon a standard environmental inventory of tritium so that future releases do not exceed this level at any time. An artificial inventory of 20 kg of tritium has been suggested by Lewis [2]. From this inventory Lewis has calculated the degree of trapping of tritium required for the safe operation of thermonuclear and nuclear fission power sources.

Table 1 summarizes some of Lewis' data. These data illustrate that nuclear fission power sources will require much less stringent housekeeping to limit tritium releases to the desired amount. However, the fission process produces many other radioactive fission products which have to be contained.

Table 1

Tritium Production for Thermonuclear Fusion and Nuclear Fission Power Sources

	Thermal/electric efficiency	T production in kg/a	Fraction released	World Inventory at equilibrium (kg of T)
2,000 GWe of thermonuclear fusion	40%	114,400	10^{-5}	20.0
	60%	76,300	10^{-5}	13.5
2,000 GWe of nuclear fission	30%	3.3 - 8.2*	0.2	11.6 - 29.0
	40%	2.5 - 6.2*	0.2	8.7 - 22.0

*Depending upon the fissioning nucleus, the tritium yield varies by about a factor of 2.5.

Albenesius [3] in 1959 first showed that tritium is produced in a small fraction of the thermal neutron fissions of ^{235}U. About one tritium atom was formed for every 10^4 fissions. Previously it had been known that ^{4}He is produced in coincidence with fission at a rate between 20 and 33 ^{4}He atoms per 10^4 fissions. (See a review by Nobles [4]). Now many light particles have been shown to be produced during the fission process. Some of these are n, ^{1}H, ^{2}H, ^{3}H, ^{3}He, ^{4}He, ^{6}He, ^{8}He, ^{10}He, Li and Be. Table 2 lists the approximate yields of light particles for the spontaneous fission of ^{252}Cf as compiled by Dudey [5].

Table 2

Light Particle Data for the Spontaneous Fission of ^{252}Cf. (According to Dudey [5])

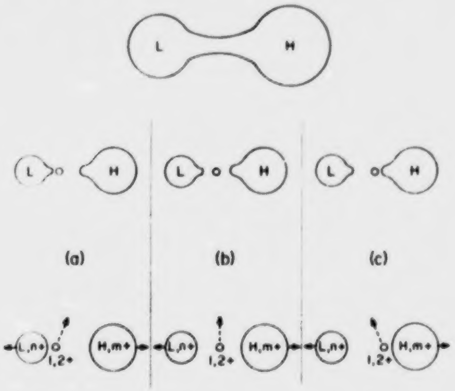
Particle	Atoms/Fission	Maximum Energy MeV	Most Probable Energy, MeV
^{1}H	5.1×10^{-5}	17	9
^{2}H	2.0×10^{-5}	18	7
^{3}H	1.9×10^{-4}	20	8
^{3}He	$<2.9 \times 10^{-5}$	31	17
^{4}He	3.3×10^{-3}	34	16
^{6}He	7.8×10^{-5}	26	13
^{8}He	5.9×10^{-5}	24	<13
^{10}He	3.0×10^{-7}	--	--
Li	3.9×10^{-6}	32	--
Be	$>3.0 \times 10^{-7}$	--	--

Theory of Light Particle Formation

From angular dependency studies it has been established [6,7] that the ^{4}He particles produced during fission (as opposed to those emitted from the radioactive decay or "boil-off" reactions of the fission fragments) are produced in the neck of the scissioning nucleus, as predicted from the liquid drop model of the fission process [8,9]. Figure 1 shows a simplified diagram of the fission process for one of the few events which results in the isolation of a small mass

Figure 1

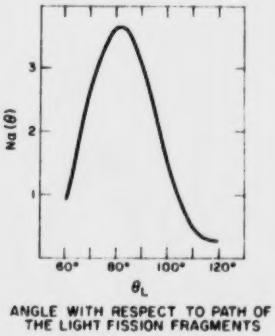
Simplified diagram of fission events which result in a third particle of Z = 1 or 2 being formed in the neck region of the scissioning nucleus. L denotes light mass fragment and H the heavy mass fragment. The charge on the fission fragments are noted as n+ and m+.



between the two main fission fragments. The light, low Z (atomic number) fragment will form between two fragments which have a high charge associated with them. The energy of the light particle will result from the coulombic repulsion of the positively charged fragments. The repulsion force will give the light fragments a preferred direction which will be nearly perpendicular to the direction of the fission fragments. Figure 2 shows the distribution of alpha

Figure 2

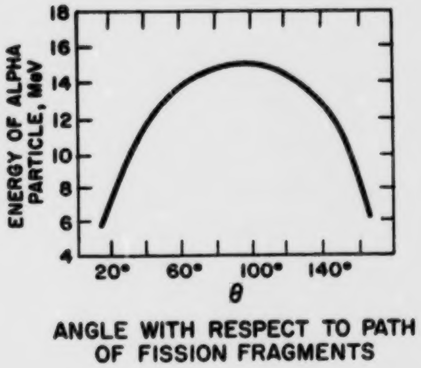
The angular dependence of the alpha particles emitted during fission process as the function of the angle with respect to the direction of the light fragment.



particles relative to the direction of the light fragment from fission. It shows a peak near 85° which is slightly different from the predicted 90° due to the greater influence of the heavy fragment with its greater charge [6]. Figure 3 shows the distribution of the energies of the alpha particles as a function of the angle relative to the direction of the fission fragments. The maximum energy occurs at an angle of 90° which corresponds to the maximum coulombic repulsion [7].

Figure 3

The distribution of the energy of fission alpha particles as the function of the angle with respect to the path of the two fission fragments.



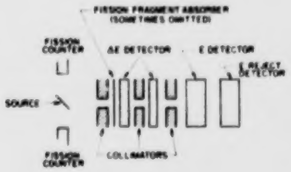
It is assumed that these same dependencies are present in the production of the other light particles, especially tritium.

Method of Measurement

Two different methods are used to measure the yield of tritium formed during the fission process. One is the counter method and the other is the chemical isolation method. The counter method

Figure 4

A counter arrangement used in the counter method for measuring energy and identifying particles emitted during the fission process.



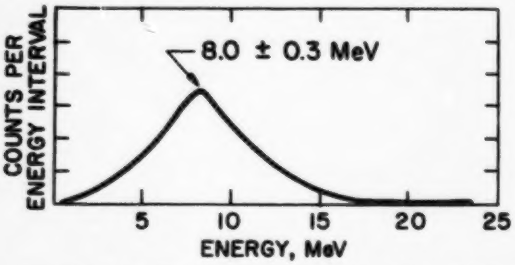
actually counts the light particles, and their yields are only known relative to one of the measured count rates. Figure 4 shows a typical counter. The type of particle is determined by its specific ionization (dE/dx) and its total energy (E) by complete stoppage in the E-detector. This method allows for measurement of all light particles at the same time but their yields are not absolute, only relative. A second measurement is necessary before the yields can be calculated. Usually the ^{4}He yields are measured in coincidence with the fission events to give the yield of ^{4}He per 10^4 fissions.

The chemical isolation method is applicable only for tritium. All other light particles are either not radioactive or have half-lives too short to allow radiochemical isolation and measurement. The tritium is isolated as tritiated water, THO, and its decay rate is measured in a liquid scintillation counter. The total fissions are determined by radiochemical assay of a fission product (i.e., ^{140}Ba) of known yield.

The energy distribution of the tritons causes difficulties in determining the yield in both methods. Figure 5 shows a typical triton energy spectrum. In the counter methods, many of the low energy tritons are undetected because they are stopped in the fission fragment absorber, stopped in the ΔE detector or even stopped in the sample (for thick samples). In the chemical isolation method it is possible that high energy tritons will escape from the sample (at least from surfaces) and will be undetected. Without precautions, these types of errors will lead to low tritium yields.

Figure 5

Distribution of the energies of the tritons produced during the spontaneous fission of ^{252}Cf.



TRITON ENERGY SPECTRUM COINCIDENT WITH FISSION OF ^{252}Cf (FROM REF. 18)

Tritium Fission Yields

Table 3 summarizes the literature data on tritium fission yields for some nuclides which are important in nuclear fission power sources. The yields of alpha particles have been shown [4] to be a function of the value of Z^2/A; where Z is the atomic number of the fissioning

Table 3

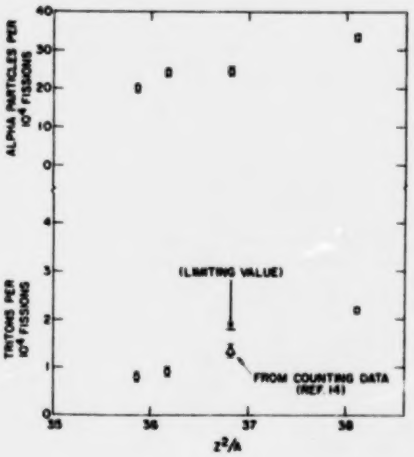
Some Tritium Fission Yield Data

Fissioning nucleus	Reaction	Method	Tritium atoms per 10^4 fissions	Refs.
^{234}U	$^{233}U + n_{th}$	chemical isolation	0.91 ± 0.06	10
		counter - dE/dx, E	0.86 ± 0.07*	11
^{236}U	$^{235}U + n_{th}$	chemical isolation	0.5 to 1.0	3
		chemical isolation	0.95 ± 0.08	12
		chemical isolation	0.80 ± 0.10	13
^{240}Pu	$^{239}Pu + n_{th}$	counter - dE/dx, E	1.35 ± 0.13**	14
		counter - dE/dx, E	1.67 ± 0.08**	15
		chemical isolation	<1.82	10
^{252}Cf	spontaneous fission	chemical isolation	2.22 ± 0.05	16
		counter - dE/dx, E	2.22 ± 0.44***	17
		counter - dE/dx, E	2.14 ± 0.07***	18

*Calculated based on 24.1 alpha particles per 10^4 fissions.
**Calculated based on 24.4 alpha particles per 10^4 fissions.
***Calculated based on 33.4 alpha particles per 10^4 fissions.

Figure 6

Plots of the yields of (a) alpha particles and (b) tritium atoms as a function of the value of Z^2/A of the fissioning nucleus showing the similarity between the dependency for the two particle yields.



nucleus and A is the mass number of the fissioning nucleus. Figure 6 has plots of alpha and tritium yields as a function of Z^2/A of the fissioning nucleus. The similar relationships suggest that there is a common mechanism of production of the tritons and alpha particles. It is probably possible to determine the tritium fission yields of other nuclides by extrapolation of the data in Figure 6.

The only tritium yields available to date are those for thermal neutron fission. Of primary importance to reactor operation is the effect of neutron energy upon the tritium yields. This information will be needed to allow for prediction of the actural amount of tritium that is to be present in reactor operation where there is a substantial fraction of the neutrons with energy above thermal energy.

Burn-up Measurements

If tritium fission yields can be determined very accurately, they could be used to determine the amount of material that has fissioned, i.e., the burn-up [19]. The determination of burn-up is very important in calculating the cost of operating a nuclear fission power reactor. Fuel costs are based on how much fuel has actually been used. Therefore, economics are closely allied to the ability to measure the burn-up. A small error in determining burn-up could mean millions of dollars. The escape of tritium from the fuel, either by high energy penetration of the tritons into the cladding material or by diffusion out of the fuel, could lead to difficulties in the determination of burn-up by this method. It can also lead to the possible contamination of the reactor coolant material.

Conclusions

Tritium is produced from fission. The consequence of this can be useful and/or hazardous. It will be very important to the future of nuclear power sources to know the actual yield of tritium so that all precautions can be exercised to limit any harmful effects. It should also be remembered that there are other sources of tritium which are more productive than the fission process: neutrons on ^{6}Li and neutrons on deuterium. Very small amounts of lithium impurity can produce much more tritium than is produced by the fission process.

ACKNOWLEDGMENT

This report is based on work performed under the auspices of the U.S. Atomic Energy Commission.

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TRITIUM PRODUCTION IN NUCLEAR REACTORS

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The basic mechanisms for generation of tritium in operation of a nuclear reactor have been reviewed in several excellent articles published during the past few years. These articles also provide the basic physical constants for the processes, and estimates of tritium generation rates. We especially mention the excellent publication by Jacobs [1], the review article by Peterson et al. [2], and the article by Ray [3].

Not much new of a fundamental nature can be added to the information provided by these articles. However, new information has begun to appear on actual measured amounts of tritium generated in operating reactors [4-7]. Furthermore, some new trends are developing in designs and modes of operation of power reactors, trends that could have a significant effect in the future on the release rates of tritium from power reactors and the places where releases occur. The following is an updating of the reviews of two to three years ago, concerned with rates of tritium production in nuclear power reactors, taking new information into account.

Basic Mechanisms

Yield from Fission: The elementary production of tritium in nuclear reactors has been reviewed by Horrocks [8]. The method of tritium generation common to all nuclear reactors is ternary fission. This is a relatively rare mode of fission, in which the fissile nucleus breaks into two heavy fragments and one light fragment, rather than the normal two heavy fragments. The light fragment can be any one of several light stable or unstable nuclei. In about 1 out of 10^4 fissions, tritium is formed as a ternary fission product. Measurements are still underway to improve knowledge of the yield for different fissile nuclei. The yield appears to vary appreciably from one fissile nuclide to another. The yield from fission of ^{239}Pu exceeds that from ^{235}U by a factor of about two. It is interesting in this connection to recall that as fuel burnup progresses in a uranium-fueled reactor, the fraction of power generated in fissioning ^{235}U diminishes, while the fraction of power generated in fissioning ^{239}Pu increases. Therefore, the rate of generation of tritium should be greater in heavily burned fuel elements from converter reactors than from elements which are fresh. This increase seems not to have been investigated until now, perhaps partly because the change in tritium production with burnup is comparable with the uncertainty in calculation of the tritium production rate.

The data pertinent to production of tritium through fission in a water-moderated reactor are listed in Table 1. A water-moderated nuclear reactor operating to produce 1,000 megawatts of electrical power would produce about 3,000 megawatts of heat, at efficiencies in the range appropriate to current water-moderated reactors. At this power level, and at the assumed tritium yield of 10^{-4} from fission, the tritium production rate would be about 1.5 grams, or about 15 KCi, of tritium per year. This tritium would be distributed among tens of thousands of fuel rods in the reactor, so that the production rate, on the average, is less than 1 Ci of tritium per rod per year.

Table 1

Features of a 1,000 MWe Reactor
Associated with Tritium Production

Electrical Power	1,000 MW
Thermal Power	3,000 MW
Fissions/a	2.8×10^{27}
Tritons/a	3.0×10^{23}
Tritium/a	1.5 g = 15 KCi

Fast breeder reactors should generally have a greater specific rate of generation of tritium by fission, because of the higher tritium yield from fission of plutonium [9]. The rate of generation per megawatt should be about double that in a water-moderated reactor.

One other kind of reactor that has been investigated extensively in this country for commercial electrical power production is the molten salt breeder [10]. The tritium produced in a molten salt breeder would be generated in the molten salt that serves as the fuel, the coolant, and which also supplies the greater part of the neutron moderation. This reactor would produce tritium from ternary fission of ^{233}U, which is the basic fuel. Recent measurements indicate that the yield from fission of ^{233}U is about the same as that from fission of ^{235}U [8].

In addition, ^{233}U is the ultimate fuel of the High Temperature Gas-Cooled Reactor, and also the seed-blanket version of the water-moderated power reactor being developed by the Naval Reactor Branch of the AEC. The specific fission yields of these reactors would be comparable to those of ^{235}U fueled reactors.

Yield from Reactions in Control Rods: The most common neutron absorber in control rods of reactors is boron. The reaction

$$^{1}_{0}n + ^{10}_{5}B \rightarrow ^{7}_{3}Li + ^{4}_{2}He$$

has a high cross section for thermal neutrons, and this property is used to provide reactivity control through insertion of fewer of more boron-containing control rods. The fast neutron reactions on the lithium created from the boron capture

$$^{1}_{0}n + ^{7}_{3}Li \rightarrow ^{1}_{0}n + ^{3}_{1}H + ^{4}_{2}He$$

lead in turn to formation of tritium. The final yield of tritium increases with time as the quantity of ^{7}Li in the control rods builds up. The rate of production of tritium in control rods depends strongly on the kind of reactor and the control rod programming strategy, because rods have to be inserted in the operating reactor core for tritium to be produced. For a typical water reactor using boron-loaded control rods, the estimated rate of tritium production in the control rods should vary from about 2 KCi per year for relatively new control rods to about 20 KCi per year for well-burned control rods.

Other Reactions in Fuel: The proposed Molten Salt Breeder Reactor (MSBR) uses a molten circulating fuel which consists of 72% LiF, 16% BeF_2, and 12% ThF_4, with a small amount of UF_4. The tritium production in a 1,000 MWe version of this reactor would be about 900 KCi/a through direct neutron reaction on the lithium. The lithium proposed for use is 99.995% ^{7}Li because the production rate of tritium would be substantially greater through thermal neutron capture if ^{6}Li were used.

Yield from Reactions in Coolant: Pressurized water reactor (PWRs) commonly use soluble boron in the coolant for reactivity shimming. The two-stage reaction by which boron exposed to a neutron flux is partly converted to tritium occurs in this case directly in the coolant. Estimates of the rate of tritium production by neutron reaction with the soluble boron vary from about 500 Ci to about 1 KCi per year, for a 1,000 MWe PWR. A small amount of tritium is produced in water reactors from direct neutron reaction of LiOH, which is sometimes used for pH control.

In a 1,000 MWe High-Temperature Gas-Cooled Reactor (HTGR) using helium as the coolant, up to 6 KCi of tritium per year would be produced by neutron reaction with the coolant. Neutron capture in deuterium takes place with a very small cross section. The rate of production of tritium from neutron capture in the small amount of deuterium normally present in the coolant of a light water reactor is very small, of the order of 10 Ci/a for a 1,000 MWe reactor. The rate of generation of tritium in the heavy water of a heavy water-moderated reactor (HWR) is substantial, amounting to about 600 KCi/a for a 1,000 MWe reactor.

Solid Shim: Some water reactors use solid neutron absorbers for reactivity shim. Principal among these are some which use boron-loaded poison curtains for reactivity holddown during the early period of operation of a new reactor. It has been estimated that about 2 KCi of tritium are generated in the poison curtains used in the first burnup cycle of a new 1,000 MWe water reactor.

Production in Different Reactor Types

The rates of production of tritium in the different regions of different kinds of reactors are summarized in Table 2. The higher rate of production of tritium in the fuel of the Liquid Metal Fueled Breeder Reactors (LMFBR) is a consequence of the use of plutonium as the fuel, rather than ^{235}U.

Table 2

Tritium Production Rate
for Various Reactor Rypes (kCi/a)

	Fuel	Control Rods	Coolant	Total
PWR	15	1	small	16
BWR	15	6	small	21
HTGR	15	undetermined	6	21
HWR	15	undetermined	600	615
LMFBR	30	undetermined	---	30
MSBR	15	undetermined	900	915

Factors Affecting Location and Amount of Release

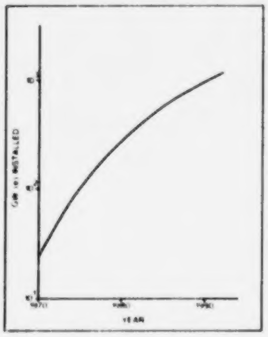
Permeability of Fuel Cladding: Most power reactors use UO_2 as the fuel, at a specific gravity of about 10. In this material, the tritium nuclei from fission have a range of only a few thousandths of an inch. The operating temperature of the uranium oxide is high, however, and the tritium vapor diffuses readily and is evolved into the gas plenum in each fuel rod. The subsequent fate of the tritium gas depends on the composition of the fuel cladding. The older practice with respect to fuel for water-moderated reactors is based on use of stanless steel cladding. Stainless steel at the elevated temperatures of reactor fuel cladding is relatively permeable to hydrogen and its isotopes. A large fraction of the tritium generated in the fuel diffuses slowly through stainless steel cladding to exchange with hydrogen in the coolant water. The newer practice is to use a zircaloy cladding for the fuel. The zircaloy demonstrates a generally superior reliability with respect to development of pin hole leaks and other defects. It is also less permeable to tritium, presumably through trapping the tritium as zirconium hydride. Over the core life of a water reactor with zircaloy clad fuel, the tritium generated by ternary fission stays for the most part in the intact cladding. We shall present some data on the degree of holdup later in this paper.

The preceding considerations refer principally to water-moderated reactors. Some other reactors have fuel elements that differ physically and chemically from elements in water reactors. High Temperature Gas-Cooled Reactors use fuel consisting of small spherules, some of enriched uranium and some of thorium, in a graphite matrix [11]. The cladding is pyrolytic graphite. The rate of tritium generation from fission is about the same in these reactors as with other reactors. There is considerably less experience on which to base estimates of the fractional release of tritium to the coolant in High-Temperature Gas-Cooled Reactors.

There is still uncertainty as to the precise cladding material that will be used for fuel in commercial-size fast breeder reactors, and so the permeability of the fuel cladding in these reactors is not

Figure 1

Projected Rate of Nuclear Power Generation



yet known. It is unlikely, however, that any material used for fast reactor fuel cladding will include the specific feature of high affinity of zirconium for hydrogen. It is, therefore, reasonable to assume that a large part of the tritium generated in the fuel of commercial fast reactors will diffuse into the coolant.

Control Rods and Solid Shim: Evidence has been accumulating that most of the tritium generated in boron-loaded control rods remains in the rods and does not diffuse into the coolant [4,5]. This seems to be so in spite of the fact that the boron is contained in stainless steel, which when used to clad fuel is relatively permeable to tritium. The reason for the difference is not clear. One contributing factor might be a lower temperature of the stainless steel cladding of control rods, which would tend to lower the rate of diffusion of tritium. Another and more likely reason might be the trapping of tritium by the lithium produced by the (n,α) reaction on ^{10}B, to form stable lithium hydride. Another possibility is formation of organic compounds through interaction with carbon freed from the boron carbide. Similarly, most of the tritium formed in the boron steel shim curtains of BWRs and the burnable poison rods of PWRs appears not to diffuse out into the coolant.

Tritium in the Coolant: In some cases the disposition of tritium in the coolant is well understood, in some cases it can be predicted with a high priority, and in some cases it can only be guessed at, because of inadequate experience. Tritium exchanges readily with the hydrogen in light water, and so it should become uniformly dispersed as ^{1}HTO in the coolant of light water-moderated reactors. Likewise, tritium in the coolant of a heavy water-moderated and cooled reactor should be dispersed as DTO.

The tritium in the molten sodium of the LMFBR would be retained in the sodium as a hydride or would be released to the cover gas, depending on the chemistry of the sodium and its additives. The experience at the Experimental Breeder Reactor-II (EBR-II) at the National Reactor Testing Site at Idaho Falls, Idaho, indicated that a substantial fraction of the tritium appears in the sodium cold traps [6].

The tritium in the helium coolant of the High-Temperature Gas-Cooled Reactor would probably be scavenged in the charcoal beds and cold traps of the coolant gas cleanup system.

In the proposed Molten Salt Breeder Reactor, the rate of escape of tritium from the molten salt could equal the rate of production after some time, although experience with the Molten Salt Reactor Experiment indicated a considerable internal holdup in the reactor.

Coolant Turnover in Water Reactors: The design of water reactors permits continuous replacement of the coolant water. The turnover in BWRs is more rapid than in PWRs, and can take place at a rate up to that corresponding to complete turnover several times each year. The turnover of coolant water leads to lower inventories in the coolant than would otherwise occur, while the difference is released in normal liquid or gaseous effluent. As a point of reference, if the coolant replacement rate corresponded to complete turnover each year, the inventory of tritium in the coolant would be only about six percent of the maximum asymptotic level without turnover. The asymptotic annual release rate would be essentially the rate of supply of tritium to the coolant, independently of the turnover rate.

The concentrations of tritium in effluent of BWRs are very low. Tritium concentrations at discharge are estimated to be about 10^{-5} times the maximum permissible drinking water concentrations [4].

Trend to Zero Release: Considerable interest has been expressed in designing and constructing power reactors to avoid release of all radioactive effluents. This practice would be more difficult for tritium than for other radioactive products, because it would require either complete retention of liquid wastes or the installation of an isotope separation facility to extract the tritium from other hydrogen isotopes. Without such a facility, operation under zero release conditions could lead in time to very high concentrations of tritium in the coolant, with correspondingly high exposure of operating personnel.

Observed Tritium Release Rates: The experience so far is primarily that obtained from operation of water reactors. By and large, the observed tritium releases are seen to bear out the qualitative and quantitative considerations that we have outlined.

The rates of discharge of tritium in liquid effluents from operating water reactors for the past four years has been summarized by Pomeroy [7]. The data are shown in Table 3.

Table 3

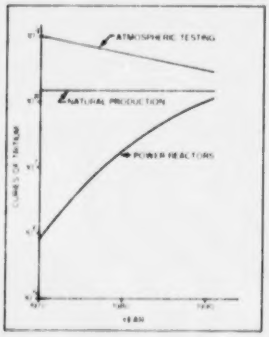
Tritium in Liquid Effluents
(Ci/a)

	1967	1968	1969	1970
Indian Point 1	300	790	1100	410
Yankee	1600	1200	1200	1500
Conn Yankee	120	1700	5200	7400
San Onofre		2400	3500	4800
Ginna				110
Big Rock Point		34	28	53
Humboldt Bay		7.2	5	7
Dresden 1		2.9	6	5
Dresden 2				31
Oyster Creek			5	22
Nine Mile Point				20

The first four reactors are PWRs with stainless steel fuel cladding, the fifth is a PWR with zircaloy cladding, and the remainder are boiling water reactors with zircaloy cladding. The effect of zircaloy in retaining the tritium is impressive. The rates of release from the boiling water reactors are consistent with the view that the tritium in the coolant is not greatly above that generated in the coolant solely through neutron capture in the small amount of deuterium in light water [4]. No detectable leakage of tritium has occurred from the fuel or the control rods. Similarly, the rate of release of tritium from the Ginna Pressurized Water Reactor shows no clear indication of tritium in the coolant greatly in excess of that produced by neutron reactions on the soluble boron used for shim [5].

Figure 2

Estimated Burden of Tritium in the Environment



Effects on Chemical Reprocessing: The use of fuel cladding that retains tritium transfers the point of potential tritium release to the chemical reprocessing plant. One chemical reprocessing plant can handle the fuel from many reactors. One of the larger fuel reprocessing plants currently planned could service about 40,000 electrical megawatts of installed nuclear power plant capacity, operated at a reasonable load factor. A reprocessing plant of this size processing fuel from water reactors would receive and liberate 600 KCi of tritium annually in spent fuel for reprocessing. Approximately four reprocessing plants will be in use in the United States at the end of the next decade: these should be the focus of attention as regards tritium releases, rather than the approximately 200 nuclear power reactors expected to be in operation at this same time.

At the time of fuel reprocessing, part of the tritium (probably less than 10 percent) remains with the pieces of chopped cladding that form the residue from acid dissoluation and are called leached hulls. This tritium with the hulls is probably retained in the form of zirconium hydride. About 20 percent of the tritium appears in the dissolver off-gas. The remainder is left in the aqueous raffinate, and is distilled off into low level waste then the high level liquid wastes are evaporated.

Some consideration could be given to operating chemical reprocessing plants on a zero tritium release basis. The need to avoid buildup of tritium activity in such an operation would argue strongly for use of an isotope separation plant.

Thermonuclear Reactors: It has been pointed out that tritium generation in thermonuclear fusion reactors will be much greater than in fission reactors. This is because the most likely fuel for thermonuclear reactors will be a mixture of deuterium and tritium, and the reactors must replace, by breeding, the tritium they will burn. On a per megawatt basis, thermonuclear reactors would generate about 10^5 times as much tritium as fission reactions [12]. The environmental problems of thermonuclear reactors will in all probability be primarily the result of tritium generation.

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TRITIUM IN PRESSURIZED WATER REACTORS

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The study of tritium in pressurized water reactors (PWRs) is interesting because tritium's half-life (12.3 years) permits long-term buildup. While its 10-day biological half-life and 18 keV β-decay energy pose no serious radiological hazard, tritium is produced in sufficient quantities in PWRs to cause concern regarding its long-term management. Since reactor coolant is recycled, tritium is retained with the plant as tritiated water and releases may occur as liquid, water vapor or hydrogen. This paper describes Westinghouse PWR Systems Division investigations into the prediction, measurement and long-term management of tritium produced in PWRs employing zircaloy-clad fuel, silver-indium-cadmium control rods, boric acid as a soluble chemical shim, and borosilicate glass in burnable poison control rods.

Predictions

Ternary Produced Tritium: The yield of tritium from the ternary fission of enriched uranium was first reported by Albenesius [1]. It was estimated that in addition to other fission products, one tritium atom was produced for every 1 to 2 X 10^4 fissions. Horrocks has reviewed this subject recently [2]. Since approximately 90 percent of the power produced in current Westinghouse PWR results from the fissioning of ^{235}U, the tritium production from ternary fission (as used in this report) corresponds to that measured by Sloth et al. [3]. This translates to a tritium production rate of 0.0105 curies of tritium per thermal megawatt day of power. The fourth column of Table 1 presents the calculated ternary fission tritium production in a PWR operating at 1,300 MWt.

Predictions for the expected release of ternary fission tritium are that 1 percent will be released from zircaloy-clad cores, and 80 percent will be released from stainless-clad cores. For purposes of plant systems design and to assure adequate design margin, Westing-

Table 1

Predicted Plant Total Tritium Production
For a Plant Operating at 1300 MWt

Cumulative Time (Effective Full Power Days)	Time Interval (Effective Full Power Days)	Average Boron Concentration (PPM)	Fuel Ternary Fission Tritium Yield (Ci)	Burnable Poison Tritium Yield (Ci)	Soluble Source Tritium Yields (Ci)
4.97	4.97	1300	68.12	5.29	5.08
11.49	6.52	1200	157.47	12.16	11.19
24.67	13.18	1120	338.11	25.82	22.76
32.42	7.75	1100	444.33	33.70	29.45
64.51	14.09	1100	637.44	47.80	41.60
49.38	2.87	1100	676.77	50.64	44.08
67.26	17.88	1100	921.82	68.03	59.49
85.68	18.42	1230	1174.30	85.51	77.22
104.88	19.20	1200	1437.40	103.31	95.26
134.98	30.10	1025	1849.90	130.44	119.44
162.52	27.54	985	2227.40	154.55	140.72
176.72	14.20	960	2422.00	166.75	151.43
197.92	21.20	900	2712.50	184.71	166.43
231.72	33.80	875	3175.80	212.75	189.66

house initially selected 30 percent of the ternary fission tritium as the amount being released from zircaloy-clad fuel. It should be noted that even if all of the tritium produced in a typical PWR were released, the discharge concentration in the plant liquid effluent would remain at a small fraction of the Maximum Permissible Concentration. To date, PWR operating experience with zircaloy-clad fuel indicates a ternary tritium release close to the predicted one percent.

Absorber Rod Sources of Tritium: Two types of neutron absorber rods are employed in a Westinghouse-designed PWR: a) burnable poison (B_2O_3) rods, used in the initial cycle to maintain a negative moderator temperature coefficient, and b) the rod cluster control rods used for reactor control. Tritium production in the burnable poison rods results from neutron reactions with boron. The neutron reactions with boron resulting in the production of tritium are:

$$^{10}B\ (n,\ 2\alpha)T$$

$$^{10}B\ (n,\ \alpha)\ ^{7}Li\ (n,\ n\alpha)T$$

$$^{11}B\ (n,T)\ ^{9}Be$$

$$^{10}B\ (n,d)\ ^{9*}Be\ (n,\ \alpha)\ ^{6}Li\ (n,\ \alpha)T$$

Figure 1 presents a plot of the energy-dependent neutron cross section for the ^{10}B (n, 2α)T reaction used in the predictions. Figure 2 presents a plot of the energy-dependent neutron cross section for the ^{7}Li (n, nα)T reaction used in the predictions. Figure 3 presents the

Figure 1

^{10}B (n,2α) T Cross-Sections (4)

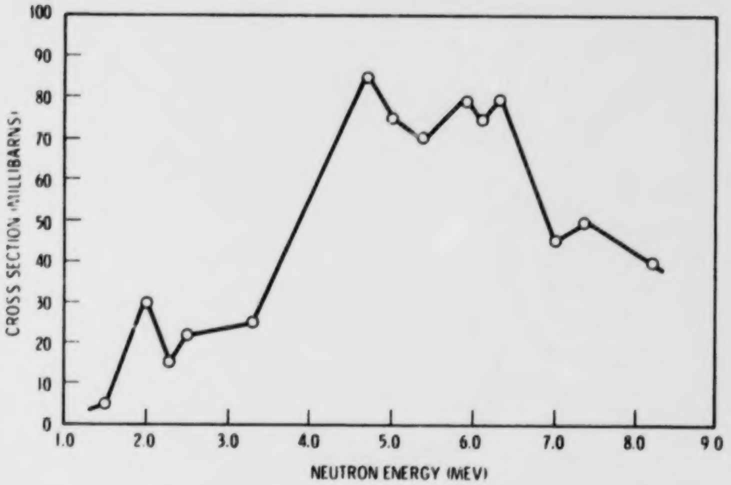


Figure 2

^{7}Li(n,nα) T Cross-Sections (4)

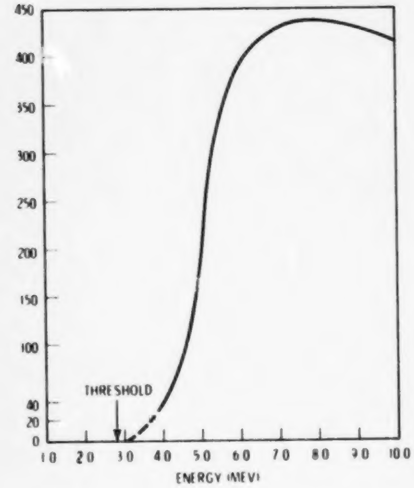
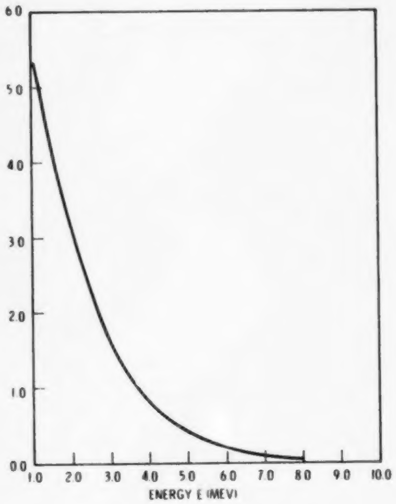


Figure 3

Core Average Fast Neutron Flux ϕ (E) Above Energy E (MeV) at 1300 MWt



core average neutron flux distribution for a Westinghouse-designed PWR operating at a power level of 1,300 MWt.

Of the above reactions, only the first two contribute significantly to tritium production in a PWR. The ^{11}B (n, T) ^{9}Be reaction has a threshold of 14 MeV and a cross section of 5 mb. Because the number of neutrons produced at this energy is less than 10^{9} n/cm^{2} sec, the amount of tritium produced from this reaction is negligible. The ^{11}B (n, d) reaction also may be neglected since $^{9*}Be$ has been found to be unstable.

Column 5 of Table 1 presents the predicted tritium production in the burnable poison rods of a reactor operating at 1,300 MWt. The burnable poison rods are removed after the first cycle and this contribution, therefore, is not present throughout the life of the plant. Such metals as Ag-In-Cd are used for the rod cluster control rods, and these metals are not a source of tritium. Current Westinghouse estimates on tritium release from stainless-clad burnable poison rods range from 30 percent to 80 percent of that produced.

Reactor Coolant System Tritium Sources: Tritium sources in the reactor coolant include neutron reactions with the soluble poison boron, the lithium used for pH control, and naturally-occurring deuterium. The ^{10}B (n, 2α)T and ^{7}Li (N, nα)T reaction cross sections are discussed later in this paper. Westinghouse-designed PWRs employ lithium for pH adjustment of the reactor coolant. A maximum level of 2.2 ppm lithium is maintained by the addition of $^{7}LiOH$ and by a cation demineralizer that will remove any excess of lithium produced in the ^{10}B (n, α) ^{7}Li reaction.

The ^{6}Li (n, α) reaction is controlled by limiting the ^{6}Li impurity in the $^{7}LiOH$ used in the reactor coolant and in the lithium from demineralizers with 99.9 atom % ^{7}Li. Westinghouse reactors have observed this limitation since 1962.

Since the amount of naturally-occurring deuterium in water is less than 0.015 atom %, the tritium produced from this reaction in PWRs is less than 1 Ci/a.

Table 2 summarizes the various spectrum-weighed neutron cross

Table 2

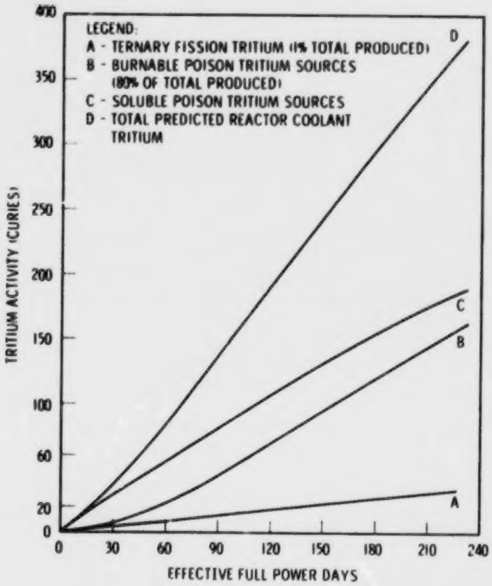
Spectrum Weighted Neutron Cross-Sections and Neutron Fluxes

Reaction	Energy	Cross-Section Barns	Core Average Neutron Flux n/cm^{2} - second
$^{10}B(n,2\alpha)T$	1 MeV $\leq$ E < 5 MeV 5 MeV > E	3.16 X 10^{-2}	4.82 X 10^{13} 5.17 X 10^{12}
$^{10}B(n,\alpha)Li^{7}$	0.625 eV $\leq$ E	3.84 X 10^{3}	2.30 X 10^{13}
$^{7}Li(n,n\alpha)T$	3 MeV $\leq$ E < 6 MeV E > 6 MeV	8.56 X 10^{-2} 4.0 X 10^{-1}	1.62 X 10^{13} 2.59 X 10^{12}
$^{6}Li(n,\alpha)T$	0.625 eV $\leq$ E	9.50 X 10^{2}	2.30 X 10^{13}
$^{2}D(n,\gamma)T$	0.625 eV $\leq$ E	4.6 X 10^{-4}	2.30 X 10^{13}

sections and neutron fluxes used in predicting activation tritium sources, whereas Column 6 of Table 1 presents the predicted reactor coolant tritium sources. Figure 4 summarizes the sources of tritium released to the reactor coolant and produced in the reactor coolant.

Figure 4

Predicted Reactor Coolant Sources of Tritium for a PWR Operating at 1300 MWt



Measurements

Zircaloy-Clad Fuel Reactor: To develop the desired data on tritium release from zircaloy-clad fuel, a program was formulated to follow the buildup of tritium at a new PWR plant. The objective was to characterize the various tritium sources and activity levels in the reactor coolant system and to provide verification of predictions and plant design criteria.

The Ginna Station of the Rochester Gas and Electric Company (RGE) was chosen for this study because it was the first large Westinghouse-designed PWR with zircaloy-clad fuel to be placed into operation in the United States. A scheme for the regular monitoring of tritium in various plant systems was developed with RGE.

The tritium log developed from the collected data was analyzed in the light of plant history to provide an understanding of the activity level behavior and its dependence on such plant operating factors as boron concentration power generated, lithium concentration, makeup rate and discharge through the waste disposal system.

The tritium activity data from the RGE samples were generated by the Westinghouse Nuclear Energy Systems chemistry department, using the procedure discussed below and obtaining samples at regular intervals from the following systems:

I. Primary System
Reactor Coolant - Hot Leg (Twice each week)
Mixed Bed Resin - Effluent (Weekly)

II. Secondary System
Steam Generator Blowdown Line (Every two weeks)
Condensate Pump Discharge

III. Boron Recycle System
Chemical Volume Control System Holdup Tank (Every two weeks)
Condensate Monitor Tank

IV. Waste Disposal System
Waste Holdup Tank (Prior to any discharge)
Condensate Monitor Tank

V. Component Cooling Surge Tank (Every two weeks)

VI. Primary Water Treatment Plant
Lake Water (Weekly)
Reactor Water Makeup Tank (Weekly)

VII. Primary Drains Tank (Every two weeks)

Each water sample received is boiled, thus, eliminating its dissolved gas content. The remaining aqueous fraction is then passed over a 6-inch column (0.5 inches in diameter) containing a mixed bed ion exchange resin in H^+ and OH^- forms. The initial 15 ml effluent fraction is discarded to prevent sample dilution, and the sample is collected at a rate of 1 ml/min. Although this effluent fraction is generally free of all radioactivity except tritium, its gamma spectrum is examined to assure radiochemical purity.

One milliliter of the effluent is mixed with 15 ml of a stock scintillator solution made from 3 parts toluene to 1 part Triton X-100. Each liter of this solution contains 150 mg of 1, 4 Bis [2-(4-Methyl-5-phenyloxazoly)] benzene (Dimethyl - POPOP) and 5.5 g of Diphenyloxazole (PPO).

It should be noted that the tritium concentration in the component cooling water reflects the use of primary plant makeup water as makeup to the Component Cooling System. The primary makeup water, being recycled via the boron recovery system, results in the retention of tritium. Tritium measurements in the secondary system were indistinguishable from distilled water blanks; the sensitivity of the analysis for tritium was 1 pCi/ml.

Supplementary data required for evaluation of these analyses are supplied by RGE on a periodic basis. These data include the makeup water and boric acid integrator readings, in addition to pertinent information on power generation.

Calculation of Tritium Buildup in the Reactor Coolant: Since the reactor coolant tritium is the sum of contributions from all the primary means of production, the contributions from the soluble poison boron and the poison rods were estimated in order to evaluate the release fraction from the fuel. The total tritium buildup in the reactor coolant system is calculated from the available data as follows:

1. Calculate the tritium inventory on the final day of a given period of power operations:

   Tritium Activity X Reactor Coolant Volume = Tritium Inventory.

2. Repeat this process for tritium inventory on the first day of the given period of power operations.

3. Tritium increase in the coolant is the difference between the inventories on the first and final days.

4. Account for tritium discharge and tritium input:

   a. Discharge = Total Makeup (Reactor Makeup Water and H_3BO_3 Additions) X Average Tritium Activity in the Reactor Coolant
   b. Input = Total Makeup X Average Tritium Activity in the Makeup Water.

5. Add the difference between Discharge and Input to increase in tritium buildup.

The above procedure was used to determine the total tritium inventory in the reactor cooling and related systems. This inventory is included in Table 3, which also lists the predicted inventory and

Table 3

Operating Plant Predicted and Observed Tritium in All Plant Liquids

Cumulative Time (Effective Full Power Days)	Time Interval (Effective Full Power Days)	Average Boron Concentration (PPM)	Predicted Tritium (Ci)	Observed Tritium (Ci)	Ratio Observed/ Predicted
4.97	4.97	1300	5.96	5.34	0.90
11.49	6.52	1200	13.89	11.18	0.80
24.67	13.18	1120	30.96	24.18	0.78
32.42	7.75	1100	41.84	32.18	0.77
46.51	14.09	1100	63.06	54.93	0.87
49.38	2.87	1100	67.58	61.29	0.91
67.26	17.88	1100	96.84	92.03	0.95
85.68	18.42	1230	130.40	124.02	0.95
104.88	19.20	1200	166.05	159.34	0.96
134.98	30.60	1025	218.11	206.98	0.95
162.52	27.54	985	267.44	246.98	0.92
176.72	14.20	960	292.19	302.68	1.04
197.92	21.20	900	328.08	361.18	1.10
231.72	33.80	875	384.36	427.58	1.11

the ratio of that observed to that predicted.

During the first several weeks, when the coolant boron concentration was highest and no other significant tritium sources existed, the observed reactor coolant tritium confirmed the soluble poison tritium predictions. Although a 10 percent divergence was noted between measurements and predictions at the end of the period, the observed tritium sources in a plant employing zircaloy-clad fuel are one-tenth that of a plant employing stainless-clad fuel.

It should be noted that during the recording of the measurements summarized in Table 3, the reactor was operating with 44 burnable poison rods. After the Table 3 data were taken, 36 of the 44 burnable poison rods were removed during the first cycle because they were no longer required to maintain the negative moderator temperature coefficient. There should be a substantial reduction in the measured plant tritium during the current operating period if the burnable poison rods contributed strongly to the tritium buildup in the reactor coolant.

Stainless-Steel-Clad Fuel Reactors: Review of the tritium data reported by Yankee Rowe reveals that, following refueling shutdowns, an uncommon increase occurs in the level of tritium in the reactor coolant. As Yankee Rowe was the only available PWR operating with recycled fuel, it was decided that tritium behavior in the Connecticut-Yankee reactor coolant would be studied during the initial operations of Core II. Since both Yankee Rowe and Connecticut-Yankee employ stainless-steel-clad fuel, it was believed that the Connecticut-Yankee refueling experience could confirm the phenomenon observed at Yankee Rowe.

The refueling shutdown at Connecticut-Yankee resulted in the removal of 48 Region I stainless-steel-clad assemblies and 4 Region III assemblies clad with zircaloy. Based on burnup figures given in the Connecticut-Yankee monthly report for May 1970, removal of those assemblies was equivalent to removal of 57.5 out of 157 average burnup assemblies, leaving 63.2 percent of the fuel to be recycled.

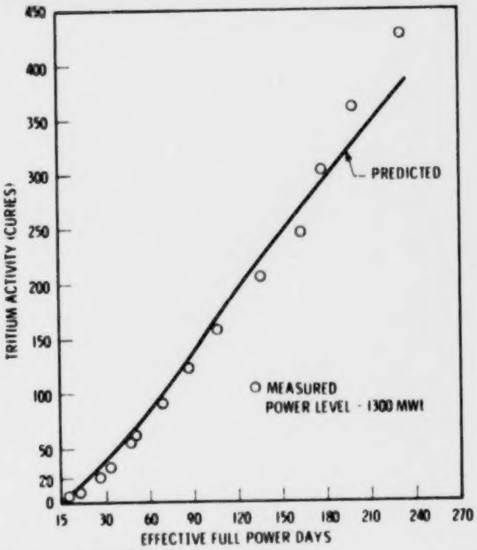
An inventory indicated that during operation two-thirds of the tritium produced in the fuel of Core I had been released through the stainless steel cladding. Based on calculations, approximately 63 percent of the remaining tritium was recycled and was available for release after startup of Core II. Of the available tritium inventory in the fuel, including that produced from Core II startup to July 23, 1970, at least 56.5 percent had been released to the coolant.

The release rate of tritium [69 Ci per effective full power days (EFPD)] during the initial startup period of Core II far exceeds the average release rate of Core I, estimated as being 13 Ci/EFPD. From subsequent tritium data derived from Connecticut-Yankee, it is apparent that the tritium buildup rate had decreased markedly to the point where the normal dilution had reduced the coolant tritium level by a factor of 2: from 7.26 µCi/ml on August 16, 1970 to 3.7 µCi/ml on September 2, 1972.

It has been confirmed that relatively large amounts of tritium were released from the Connecticut-Yankee Core within the first few weeks of operation. The release rate was seen to respond sharply to reactor operation at power levels of close to 100 percent, and would then taper off after about 2 weeks. During this entire period, the reactor coolant boron concentration remained essentially constant. The large release of tritium experienced following a refueling shutdown is attributed to a redistribution of power and temperature in

Figure 5

Reactor Coolant Tritium Sources Measured and Predicted

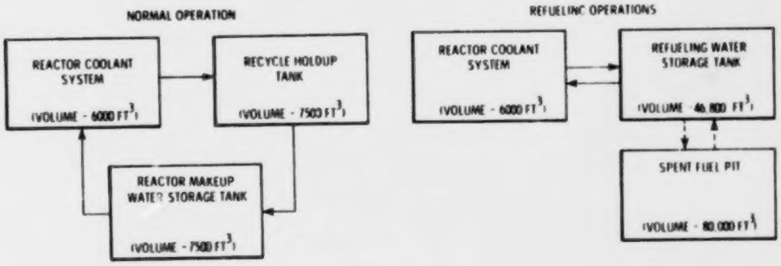


the recycled fuel assemblies.

Recently, a partial refueling was accomplished at the RGE plant, which employs zircaloy-clad fuel. During the first month of reactor operation after refueling, no increase in tritium release from the core was observed.

Figure 6

Flow Paths for Mixing Reactor Coolant During Normal Operation and During Refueling



Long-Term Buildup in Plant Retaining Tritium

Based on the assumption that all of the tritium in the plant liquid will remain within the plant during its entire operating life, an analysis was performed to establish plant operating conditions. The distribution of the prediced tritium sources during normal operation and during refueling is shown in Figure 6. No mixing between the spent fuel pit water and refueling water was assumed for this study. However, actual plant operating experience indicates that some mixing does occur, thus, providing additional dilution volume.

Figure 7 illustrates the buildup of tritium in the reactor coolant over a 32-year-period (equilibrium), assuming a plant operat-

Figure 7

Reactor Coolant Tritium Buildup - Zero Losses
1300 MWt Reactor

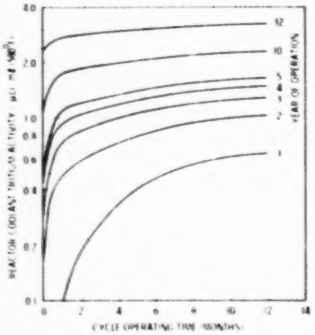
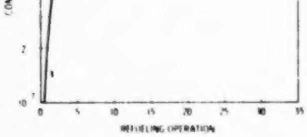


Figure 8

Containment Air Tritium Activity During Refueling
Operations - Zero Losses 2 Loop Reactor



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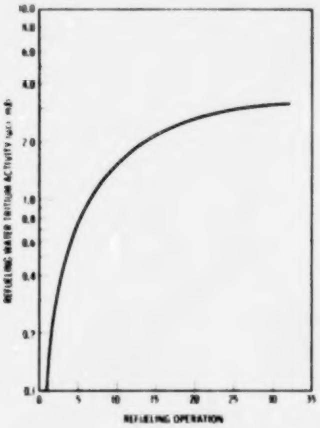
ing power of 1,875 MWt and no discharge of tritium. The maximum expected reactor coolant tritium activity is 3.3 μCi/ml (4.5 μCi/g) of coolant, which occurs after approximately 32 years of operation. Currently, PWRs with stainless-steel-clad cores have operated satisfactorily with coolant tritium levels in excess of 5 μCi/g of coolant.

Figure 8 illustrates the tritium activity in the containment air during refueling. Figure 9 illustrates the buildup of tritium activity in refueling water over the life of the plant, again assuming no tritium losses and no mixing with the spent fuel pit.

Figure 9

Refueling Water Tritium Activity Buildup - Zero Losses 2 Loop Reactor



The results of these studies clearly indicate that a PWR employing chemical shim, silver-indium-cadmium control rods and zircaloy-clad fuel, has the potential to be operated for its entire life without any need for intentional tritium dumping. From a practical point of view, it is anticipated that actual plant tritium losses will occur during containment purging and from unrecoverable plant leakage. These losses (expected to be <100 Ci/a) should remove any plant operating restrictions imposed by recycling tritium.

Conclusions

1. Reaction cross sections for $^{10}B\ (n, 2\alpha)T$ were verified.
2. Gettering ability of zircaloy for tritium was verified.
3. For two plants similar except for core clad material, tritium observed in the coolant from a plant employing zircaloy clad was less than 10 percent of that from a stainless-clad plant (430 Ci versus 4,609 Ci).
4. Additional research is required to verify the release rate from stainless-clad absorbers.

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TRITIUM EXPERIENCE IN BOILING WATER REACTORS

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Tritium associated with operation of boiling water reactors (BWRs) supplied by the Nuclear Energy Division of the General Electric Company has been observed over the past decade. Initial calculations were made in 1960 [1]. Five other summary-type reports of experience were presented during the period [2-6]. This paper updates the earlier reports.

Tritium is the only radioisotope of hydrogen and has a mass number of 3. Other isotopes of hydrogen are nonradioactive, and are ordinary hydrogen (protium) and deuterium. The tritium radioactive half-life is about 12 years. It emits no gamma radiation, but emits a beta particle of very low energy (0.018 MeV maximum, 0.006 MeV average) as it decays to helium-3. If the accumulation of tritium in the reactor system is at a location of the significant core neutron flux, decay to helium-3 is counterbalanced by the significant cross section of 5,400 barns for helium-3 to form tritium again [7]. Tritium can exist as a gas, but its most likely form in a water reactor is just as one or two of the atoms of hydrogen in water.

Principal Sources of Tritium in a BWR

In a water-moderated and cooled reactor, tritium appears from three principal sources: activation of the naturally occurring deuterium, as a product of fission, and as a product of the use of boron for reactivity control. Due to the relatively low concentrations of tritium found in BWR process fluids, there is some uncertainty as to the contributions appearing in reactor water from the various possible sources.

Deuterium Source: An obvious method of forming tritium is neutron bombardment of the $\sim$150 ppm of deuterium which occurs in natural water. The formation of additional deuterium by neutron bombardment of ordinary hydrogen is usually a minor source compared to the $\sim$150 ppm of deuterium present natually in ordinary water. To double the deuterium concentration in ordinary water would require an average reactor water irradiation age of about a year in a water reactor. The tritium formation reaction has a cross section of 5.7×10^{-4} barn.

The present design basis estimate for tritium from the deuterium source in a GE-BWR of current design is calculated to be $(1.3 \pm 0.4) \times 10^{-4}$ μCi/sec MWt. The uncertainty indicated is derived from the estimated errors in selecting values for the coolant volume and density, the abundance of deuterium present as a function of estimated typical reactor water age, and the profile of the neutron flux in and around the reactor core.

An independent audit on the deuterium source was provided by the USPHS studies [10] at Dresden 1 in 1968. The nominal reactor power level is 700 MWt. The calculated formation rate was 0.06 μCi/sec. Based on the concentration in reactor water and the calculated turnover time of plant water, the measured appearance rate in reactor water was 0.05 μCi/sec. Thus, the Dresden 1 tritium in reactor water at that time apparently could be accounted for by activation of deuterium.

Fission Source: In 1959, Albenesius [11] reported that tritium was a product of fission with a fission yield of about 0.01 percent. In 1968, Dudey [12] estimated fission yields from thermal neutrons as 0.013 percent for ^{235}U and 0.023 percent for ^{239}Pu. Both sources are present in water reactor fuel, and, thus, the tritium formation rate in fuel is probably of the order of 0.25 μCi/sec·MWt.

Of significance in water reactor operation is the "appearance" rate in reactor water of tritium transferring from fuel by various mechanisms. One approach to estimating this transfer rate would be to consider that intact zircaloy-clad fuel effectively prevents tritium transfer except to the extent permitted by the presence of cladding defects, and that tritium diffuses through defects in a manner similar to experience with the easily measurable noble gases. At design basis noble gas rates, the associated tritium "appearance" rate in reactor water would be of the order of 0.1 μCi/sec. For a typical large BWR (1,100 MWe $\sim$3,300 MWt), the fission source appearance rate in reactor water would be of the order of 3×10^{-4} μCi/sec·MWt on this diffusion approach, and less to the extent that noble gas diffusion was below the design basis at any time. As this appearance rate example is about twice the formation rate from deuterium, its contribution should be detectable with careful evaluation of reactor water age, etc.

Each fuel assembly is operated about four years prior to removal at refueling, and the fuel in each individual tube should contain about two curies of tritium at that time. Thus, the average for the core at any time should be about half of this, or about one curie of fission tritium per tube.

Since the fission formation rate is about 2,000 times the expected formation rate from deuterium, any large scale transfer of the fission source to reactor water should be readily detectable.

Boron Source: In a GE-BWR, reactivity control is primarily by boron in the control blades. Each cruciform blade contains 84 sealed stainless steel 3/16-inch diameter tubes which are about percent filled with boron carbide powder (with spacers providing providing proper distribution) with about 35 percent of the encapsulated space reserved to contain the gases generated.

In some plants, to control reactivity during operation of a new core, fixed position stainless steel-boron alloy "curtains" have been used. In current designs these are not required, as burnable reactivity control material within the fuel of certain fuel tubes is used for this purpose.

Boron in solution in the reactor water is not used. A redundant "standby liquid control system" containing boron solution is available but has no normal operations function. Reactor water is analyzed periodically to insure the absence of any significant boron content, which serves as a check on any unscheduled leakage from any boron source.

Several (primarily fast) neutron reactions contribute to tritium formation from boron. The more significant are believed to be:

Reaction	Fast Cross Section
${}^{7}Li + n \rightarrow {}^{5}He + {}^{3}H$	55 mb
${}^{7}Li + n \rightarrow n + {}^{4}He + {}^{3}H$	330 mb
${}^{10}B + n \rightarrow {}^{8}Be + {}^{3}H$	42 mb
${}^{11}B + n \rightarrow {}^{9}Be + {}^{3}H$	15 mb

where the lithium-7 is available from the basic reactivity control reaction:

$${}^{10}B + n \rightarrow {}^{4}He + {}^{7}Li$$

From these reactions, when on the average during the year the control elements are used to hold the order of 5 percent excess reactivity, the tritium formation from boron will be of the order of 0.1 μCi/sec·MWt.

An individual tube of control powder should contain (on the average, depending on blade usage for control) about 0.4 Ci of tritium per year of operation, or about 6 Ci after a nominal 15-year blade life.

Since the tritium formation rate from boron is estimated to be about 700 times greater than the formation rate from deuterium in reactor water, any large scale transfer of the tritium from boron should be readily detectable in reactor water.

Separate identity of any small fraction leakage of tritium to reactor water from the boron sites has not been achieved, either from the control blades or the temporary control curtains during periods of their presence.

Summary Estimate of Sources: For the example large GE-BWR (~1,100 MWe, ~3,300 MWt) the estimated tritium formation rates, and "appearance" rates in reactor water, are:

Source	Formation Rate μCi/sec	Appearance Rate in Reactor Water μCi/sec
Deuterium	0.43	0.43
Fission	820.0	0.1
Boron	330.0	?

Tritium Pathways in BWR: Tritium appearing in the reactor water is expected to be present as tritiated water. Thus, no variations in tritium concentration in the phase changes from reactor water to steam to condensate would be expected in an open cycle reactor. This has been confirmed by condensate analysis. Most liquid waste collection in the process buildings originates from reactor and power cycle equipment, so that tritium concentrations in liquid effluent would differ from reactor water only to the extent of non-process dilution source additions such as floor hosing water, main condenser tube inleakage, laundry and miscellaneous decontamination solutions, etc. Condensate storage received treated water recycling from the radioactive waste treatment system and reject water from the condensate system. Thus, all power cycle water will have a common tritium concentration.

The process off-gas systems such as the glad seal and the air ejector systems provide a chronic removal route for tritiated reactor water as humidity. Unscheduled releases of tritium to atmosphere result from process water and steam leakage to ventilated spaces, with routine releases due to evaporation from sumps, tanks and pools containing process water.

Those plants equipped to recombine radiolytic decomposition gases in the air ejector exhaust stream return the water formed and most of the humidity from the stream to the main condenser. Also some plants in current design will not use reactor steam for the turbine gland seals. Such process modifications reduce purposeful tritium releases in airborne effluents, and the recycling will increase process fluid concentrations.

Liquid waste treatment systems are also progressing in the direction of increasing decontamination of wastes (for radioisotopes other than tritium) with recycling for continued process use of the treated water. This also will cause an increase in the equilibrium concentration of tritium in process fluids.

For any water reactor system, the tritium "appearance" rate in the reactor water and the release from the plant will be in balance eventually. When recycling is maximized, equilibrium will be attained by any purposeful liquid releases, use of process water for cementing solid waste in containers, with the remainder of the "appearance" rate transferring to atmosphere from process leaks and evaporation from tanks and pools containing process water. Reduction due to radioactive decay at locations of low neutron flux should be negligible in the overall balance.

As an example for currently operating BWRs, the USPHS study [10] at Dresden 1 estimated that about 90 percent of the tritium release was in liquid effluents and about 10 percent in airborne effluents. As recycling at any plant is increased, the balance should move in the direction of airborne effluents. This will tend to reduce the already insignificant off-plant radiation dose, as the atmosphere has a far greater immediate dispersal capacity than the typical plant location waterway.

Tritium Monitoring in BWR: Sampling of reactor water for tritium in the initial period of reactor operation is appropriate to establish the chronic levels. Continued routine sampling is worthwhile if the chronic amount of tritium in the reactor water is small compared to additional stored sources which could be released from leaking fuel or control elements. An individual fuel tube will contain 0.1 Ci of fission tritium after a few months and about 2 Ci at the end of fuel life. An individual BWR control tube in a control blade will contain about 0.01 Ci of tritium from boron reactions after a few weeks operation, and about 6 Ci after a fifteen-year life. In the first cycle of BWR operation, where used, a control curtain will contain about 0.1 Ci of tritium from boron reactions after a few days operation, and about 20 Ci of the end of its useful life.

For an example, a large BWR, with a tritium appearance rate in reactor water of the order of 0.5 µCi/sec, and an average bulk process fluid age of the order of months (rather than weeks or years), the tritium content of process fluids would be the order of 10 Ci and the concentration would be the order of 10 nCi/ml. Thus, complete release from a number of fuel tubes or control elements would be required in order to change the inventory or concentration by an order of magnitude. Monthly sampling of reactor water or condensate appears appropriate to monitor for leakage from the stored sources. As long as the chronic levels in reactor water remain at the expected low level, there would seem to be no technical need to monitor liquid waste routinely for tritium, as the expected discharge should be only several curies per month.

The discharge via liquid or airborne effluents will result in off-plant concentrations many orders of magnitude below continuous exposure permissible concentrations. Thus, monitoring tritium releases is basically an auditing function. Knowledge of liquid concentration and volume discharged as liquid effluent is directly attainable. Calculation of tritium discharged to atmosphere can be made from results of cold trap samples of humidity in the exhaust streams and knowledge of air flow.

In any tritium monitoring program, sources other than reactor plant origin should be considered. In the past decade, weapons testing has contributed measurable quantities, primarily in surface waters. As most of the current nuclear power cycle produced tritium may be discharged from fuel reprocessing operations, any contribution from such adjacent facilities may influence low level detection analyses.

Early Experience - Tritium in BWR: While detailed auditing of tritium has been carried out in recent years, representative samples were taken in the earlier years of operation to evaluate the general level of insignificance. A summary of representative samples of BWR process fluids shows:

Plant	Year	nCi/ml	Cladding
Vallecitos (California)	1963	50	Mixed
Dresden 1 (Illinois)	1964	20	Zr, SS
	1965	3-7	Zr
Humboldt Bay (California)	1965	30	SS
Big Rock Point (Michigan)	1965	30	SS
Garigliano (Italy)	1965	0.5	Zr
Gundremmingen (Germany)	1967	1.5	Zr
Dresden 1 (Illinois)	1968	0.9-1.7	Zr
Humboldt Bay (California)	1968-1969	4-8	Zr

The samples generally indicated that deuterium activation was probably not the only tritium source, and that minor fractions of the larger sources, either fission produced in the fuel or boron produced in the control rods, must be escaping to reactor water. On the basis of the much higher material temperatures for hydrogen gas diffusion, and the presence of perhaps many minor and, in some of the reactors listed, a few major defects in cladding as operation continued, the fission source seems more likely than the control rod source. The 1964 Dresden sample indicated that perhaps one percent of the fission-produced tritium escaped from the fuel. At the time of sampling about a fifth of the Dresden core was stainless-steel-clad and the remainder zircaloy. Some major cladding defects were present, but were mostly in the stainless steel.

The 1965 Dresden samples were taken after the removal of the stainless-steel-clad fuel from the core, and at a time when no significant cladding defects were present. The 1965 samples indicate that perhaps 0.1 percent of the fission tritium was escaping to reactor water from the zircaloy-clad core. The 1968 samples were taken after several years of all-zircaloy cladding operation.

The Humboldt Bay and Big Rock Point samples were taken when the cores were stainless-steel-clad and some cladding defects were present. They generally indicate that the order of one percent of the fission tritium was escaping to reactor water. The Garigliano plant has used only zirconium-clad fuel and has experienced no significant cladding defects. It indicates a fission tritium escape to reactor water of less than 0.1 percent. From this earlier work, it was generally concluded that tritium diffusion through intact zircaloy cladding was minimal.

The experience did not include operation with the temporary control curtains, prior to Gundremmingen operation, and later use in Humboldt Bay in 1968. In addition to the 1967 Gundremmingen results, samples in July 1968 after use of curtains for over a year showed 3 nCi/ml in reactor water and 2 nCi/ml in liquid effluents. A concurrent sample of the Donau (Danube) River water upstream from Gundremmingen showed 6 pCi/ml, presumably of weapons testing origin.

A sample at Humboldt Bay in October 1968, just before a scheduled shutdown and with curtains in use for about a year showed 4 nCi/ml. While it is not possible to sort out leakage from the fission or the various boron sources, no significant increases were indicated due to the presence of the temporary control curtains.

To indicate the level of transfer of tritium from reactor water to reactor building air, at the Humboldt Bay plant on September 21, 1965 after scheduled reactor shutdown and removal of the reactor vessel head, PG&E sampled the air above the open reactor with a calibrated sampler and cold trap. Analysis by Hazelton showed that the concentration of tritium in the air was about 40 fCi/cm^3. This is less than one percent of the breathing limit for continuous occupational exposure all year. The reactor water concentration at the time was 30 nCi/ml.

USPHS Studies - Dresden 1 - 1968: A summary of the tritium findings in the USPHS study [10] showed:

·Reactor water	1.3-1.7 nCi/ml
·Fuel pool	0.88 nCi/ml
·Recycled waste	0.93 nCi/ml
·To stack, 6 nCi/sec	(∿0.2 Ci/a)
·To river, 50 nCi/sec	(∿2.0 Ci/a)

The tritium present could be accounted for from deuterium activation, with leakage from the larger stored sources apparently quite small. This appears compatible with indications of zircaloy cladding integrity as indicated by noble gas emission, as the 1967-68 annual averages were lower than in the three previous years.

Recent Large BWR Experience: Representative samples from first-year operation of recent large BWRs indicate:

Plant	Period	nCi/ml
Nine Mile Point (New York)	1970	0.9
Dresden 2 (Illinois)	1970	0.2-1.0
Millstone Point (Connecticut)	1971	0.6-0.9
Monticello (Minnesota)	1971	0.6-1.1

A number of sample results are available to indicate the general relationship between tritium in reactor water, liquid wastes, and stack drainage:

Reactor Water nCi/ml	Other Process Fluid Sample	nCi/ml
0.86	Radwaste	0.34
0.71	Radwaste	0.45
0.67	Radwaste	0.55
1.0	Radwaste	0.98
0.84	Stack drain	0.16

From these recent analyses, liquid waste discharge concentrations show that most of the effluent was reactor water at one time, with some dilution due to waste system collection of sources not of process fluid origin.

Oyster Creek - 1970 Experience: Oyster Creek and later plants are routinely measuring tritium in liquid effluents, and reports [13] for 1970 indicate a release of 22 curies. Based on the operating history through 1970, the tritium sources calculated to be available were:

Source	Curies	Location
Deuterium	∿5	Reactor water
Fission	∿6000	In fuel
Boron	∿2000	In control elements

By inference, the transfer of perhaps a few tenths of a percent of the fission and/or boron sources to reactor water is indicated.

Liquid Effluent Audit: In recent years, more detailed audits have been made of tritium releases in liquid effluents from domestic plants. Based on data supplied by plant operators to the USAEC Division of Compliance, as reported recently [14], the record shows for boiling water reactors:

Plant	Curies Released 1968	 1969	 1970
Dresden 1 (Illinois)	3	6	5
Humboldt Bay (California)	7	5	7
Big Rock Point (Michigan)	34	28	53
Oyster Creek (New Jersey)	--	5	22
Nine Mile Point (New York)	--	--	20
Dresden 2 (Illinois)	--	--	31

To put these reported releases in perspective, the Oyster Creek release for 1970 resulted in a plant added concentration of about 23 fCi/ml in the plant discharge canal. For comparison, the general public drinking water permissible concentration for tritium is 3 nCi/ml.

Gundremmingen Plant: The most complete study of tritium in any BWR has taken place at the Gundremmingen plant under the direction of Dr. H. J. Schroeder [15]. Extensive analyses are available of the coolant (analyses at weekly intervals since January of 1968) combined with the reasonably complete plant water discharge records. Because of its long half-life and its complete exchange with the system's water inventory, the release of this nuclide can be best determined by the total plant release rather than attempting to measure the release from the core. The data indicate that the release rate can be correlated with the total MWtD of fuel exposure (or equivalent control rod exposure) as shown by the following:

GWtD	Curies/a	10^{-5} Curies/a·MWtD
275	20.4	7.4
390	29.2	7.5
608	43.8	7.2
630	47.5	7.5

This correlation implies that a constant fraction of the total inventory of tritium is escaping per unit time, but this inventory could be either that of the fuel or that of the control rods, or perhaps both. If it is assumed to be the fuel inventory, an escape rate coefficient of 2.2×10^{-10} per second would be calculated, which indicates that the equilibrium release rate would be 11% of the production rate. Since the fuel exposure time is limited to about 4 years, the maximum release rate would be about 2.2% of the production rate, and the average release rate would be the order of one percent of production.

This estimate of the tritium release does not include the amount of this nuclide released via the 250,000 m^3/h of ventilation air flow. Calculations show that the amount of moisture carried out of the plant by this route is considerable, of the order of 100 tonnes per day. The few measurements available on the tritium content of the moisture indicate that as much as one-tenth to one-third of this material is process water. This amount is comparable to the 10 to 30 tonnes per day discharged from the liquid system. Thus, the tritium released in the liquid and airborne effluents may be comparable, but further study is required.

Significance in Effluents: Reactor water is eventually released to the environs in either the liquid or airborne effluent streams or via ventilation exhausts. None of the waste treatments in use or feasible (which significantly reduce other radioisotopes) will have any effect on the tritium content of water. The 12-year half-life of tritium dictates that it makes no practical difference whether the reactor water is disposed of immediately or after months of recirculation, storage and decay, or reuse in the plant.

Significance in effluents is equivalent to simple continuous disposal of the tritium formed or appearing in the reactor water. Any accumulation requiring periodic batch disposal is actually less desirable than continuous release, as some of the advantage of the time-dependent natural diversity factors is lost.

Based on operating experience, a zirconium-clad fueled BWR will have available, in the reactor water, the deuterium activation source and a small fraction of the fission source, for a total available rate of about 0.5 μCi/sec for the example large BWR, and the continuous emission of this rate in effluents provides an estimate of the effects on the environs.

Liquid Effluents: Less than 100 percent of the tritium appearing in the reactor water will be discharged in liquid waste. The significance of the emission will depend on the flow into which the material is released. This is usually the condenser cooling water flow, which for estimating purposes, may be taken as about 0.8 gallon per minute per electrical kilowatt. Thus, the discharge canal flow from an ~3,300 MWt plant would be about 900,000 gpm (56 m^3/sec). For a plant using cooling towers, or other forms of cooling water recirculation, the cooling system blowdown and other miscellaneous minor streams usually provide a plant flow to the waterway of about 5 percent of a once-through cooling system or about 2.8 m^3/sec.

The total appearance rate of about 0.5 μCi/sec in reactor water, therefore, could not produce an average discharge canal concentration in excess of about 10 fCi/ml for a cooling system or about 200 fCi/ml for a cooling tower plant. If the canal discharges to a small river of 3,000 cubic feet per second annual average flow, the concentration of plant origin at possible downstream water use points would be about 6 fCi/ml, independent of the type of plant cooling water system. Thus, continual drinking of downstream water all year would result in a dose of about one microrem per year, or less than 0.001 percent of natural background radiation. For comparison, in recent years fresh surface waters and milk have contained about 1 pCi/ml from weapons testing origin which produces a dose in the range of 100-200 microrems per year.

Airborne Effluents: Leakage of process fluids from water reactors into adjacent air spaces will cause tritium contamination in such air spaces. For example, the leakage of reactor water into the non-ventilated primary containment volume of a water reactor will cause tritium contamination of the containment atmosphere to the extent limited by 100 percent relative humidity in the containment volume. A

BWR primary containment drywell with a volume of about 150,000 cubic feet maintained at 125°F can hold about 1,000 pounds of water vapor to saturate. As this is a small fraction of the reactor water, a small leak could easily release this quantity over the months of a typical operating cycle; any leakage in excess of this quantity would condense and enter the liquid waste system via the drains provided. With an example BWR reactor water tritium concentration of 10 nCi/ml, the equilibrium containment atmosphere inventory would be about 5 millicuries of tritium, and would result in an atmospheric concentration of about 1 pCi/cm^3. After shutdown, if this small quantity is purged to atmosphere over a several-hour period (10,000 seconds assumed), the emission rate to atmosphere would be about 0.5 µCi/sec. Even if the wind persisted in one direction for the several hours (an unlikely event) the downwind concentration (from an assumed roof level release) would not exceed about 0.5 aCi/cm^3. As the general public breathing limit is 0.2 pCi/cm^3, a three-hour exposure during the postulated release would result in a dose of less than 0.4 nrem. The natural background dose in a three-hour period is about 40 µrem. If one wishes to postulate exposure of personnel in the drywell for an hour in the concentration of 1 pCi/cm^3 without benefit of respiratory and transpiration protection, the external dose would be about 1 µrem and the internal dose would be about 10 µrem.

Steam leaks from an open-cycle BWR will release tritium to the turbine building air. It is postulated that a steam leak of 0.1 percent of full-power steam flow (a leak of 10,000 pounds per hour) would be considered a very serious leak from the operational viewpoint, independent of radiological considerations. As the tritium content of steam will be the same as reactor water, the leakage rate of tritium would be about 10 µCi/sec. If the turbine building volume is the order of a million cubic feet with three air changes per hour, the tritium concentration would average about 0.3 pCi/cm^3. Thus, the dose rate to any personnel in the turbine building would be about 0.1 mrem per hour from continuous inhalation and much less from external radiation. The emission of this tritium in turbine building exhaust at roof level would result in an off-plant dose rate of about 10 nrem per hour.

While more of the tritium in reactor water is probably discharged in the liquid effluent, some is discharged in airborne waste streams. Forms include water vapor in process off-gas streams, steam leaks to ventilated spaces, and reactor water leakage flashing to steam in ventilated spaces. If one assumes that half of the tritium in reactor water is released to atmosphere, the example emission would be about 0.25 µCi/sec. For an elevated release to atmosphere of proper height (e.g., twice the height of nearby structures) the annual average atmospheric diffusion results in an effective atmospheric dilution flow in the range of 10^7 to 10^8 m^3/sec dependent on site conditions. Thus, the example annual average off-plant tritium concentrations in air would be the order of 10^{-2} to 10^{-3} aCi/cm^3. If discharge is at roof level, rather than elevated, the off-plant concentrations would be perhaps an order of magnitude higher as averaged over the year. These example concentrations may be compared to the 10CFR20 breathing limit for the general public of 2 pCi/cm^3, a limit which is correct only if the exposure by inhalation is continuous for the entire year. It can be seen that examples at either release height will result in an insignificant off-plant dose.

Conclusions and Expectations: Due to the low concentrations of tritium in BWR plant process fluids, and indications that only minor fractions of the inventory in fuel and/or control elements appear in reactor water, information on release rates is not conclusive. While some of the data indicate that the source is largely accounted for by activation of deuterium, the results from Gundremmingen and Oyster Creek indicate that this source amounts to only 10-20 percent of that in effluents, with the remainder possibly correlatable with the integrated power of the core. If the indication that the long-term release in either liquid or airborne effluents may be as high as the order of one percent of the fission source, then effects may be an order of magnitude higher than the examples evaluated herein. For a BWR, the conclusion that the in-plant or off-site radiation dose due to tritium is insignificant remains unchanged.

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TRITIUM PRODUCTION IN PLOWSHARE APPLICATIONS

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Detonation of a nuclear explosive produces substantial amounts of radioactive materials. A significant component of this material is tritium, and its production has been the subject of continuing research and evaluation within the Plowshare Program. A major goal of the program has been to reduce, as much as possible, the amount of tritium formed, while developing an economically attractive explosive system for industrial applications.

With the current emphasis of Plowshare shifted from excavation to underground engineering, tritium has become an even more important consideration. In natural gas stimulation and storage projects, for example, some tritium could exchange with hydrogen in methane fuel.

Sources of Tritium

Tritium is produced from two major sources: the explosive itself, and the material and rock surrounding the explosive. Tritium from the explosive comes from the fuel in both fission and fusion explosives. About 5 kCi to 20 kCi of tritium can be produced per kiloton by the fusion reaction, whereas only about 1 Ci/kg is produced in the fission process.

In the fusion process, tritium can be produced from a number of different reactions [1]. An example of such a reaction is the D(D,p)T reaction, in which two deuterium atoms fuse to produce a proton and a tritium atom. The D(T,α)n is a fusion reaction yielding a 14-MeV neutron, and in a fusion explosive, residual tritium is produced when such neutrons are captured by lithium in the explosive. In the fission process in both a reactor and a nuclear explosive, tritium is produced from ternary fission. Normally there are two fission fragments and something more than two neutrons produced per fission. In addition about 1 triton is formed for every 10,000 fissions as a result of ternary fission.

The tritium produced externally to the explosive comes primarily from neutron-capture reactions with boron and lithium. Boron-10 may undergo a $^{10}B(n,T)2\alpha$ reaction if the neutron energy is greater than 1.5 MeV. Lithium-6 may undergo a $^{6}Li(n,\alpha)T$ reaction; this reaction does not have a neutron energy threshold and thus occurs with very low- as well as high-energy neutrons. Other tritium-producing neutron-capture reactions, i.e., D(n,γ)T, are normally insignificant when compared with the tritium production from boron-10 and/or lithium-6.

About 2×10^{23} n/kt escape both from an unshielded fission explosive and from a fusion explosive. Although the fusion explosive produces more neutrons than the fission explosive, most of these neutrons are moderated significantly and over 80 percent of the fusion neutrons are absorbed within the explosive. The fusion explosive produces neutrons with energies up to 14 MeV, whereas the fission neutrons have an energy spectrum peaking at about 1 MeV, with most neutrons below 4 MeV. In general, the amount of shielding material required to capture a given number of neutrons increases with the neutron energy. Thus, less shielding is required to capture the same fraction of fission-energy neutrons as 14 MeV neutrons. Even though in a fusion explosive only a small fraction of the escaping neutrons is 14 MeV, more shielding is required around a fusion explosive than a fission explosive to achieve the same level of tritium production in the rock. By the incorporation of shielding internal and/or external to the explosive, the neutron output from the explosive, and hence the tritium formed from capture by lithium in the rock, may be reduced.

The total amount of tritium produced, and the relative contribution from each of the sources for any application, would then depend primarily upon the type and yield of the explosive and upon the composition of the material and rock surrounding the explosive. When a fission explosive is used and boron-containing shielding (i.e., boric acid, boron-loaded polyethylene, or boron carbide) is not used, almost all the tritium produced will come from neutron capture by lithium in the rock. This is the case even when the amount of lithium present in the rock is only a few parts per million (ppm) by weight.

Plowshare Applications

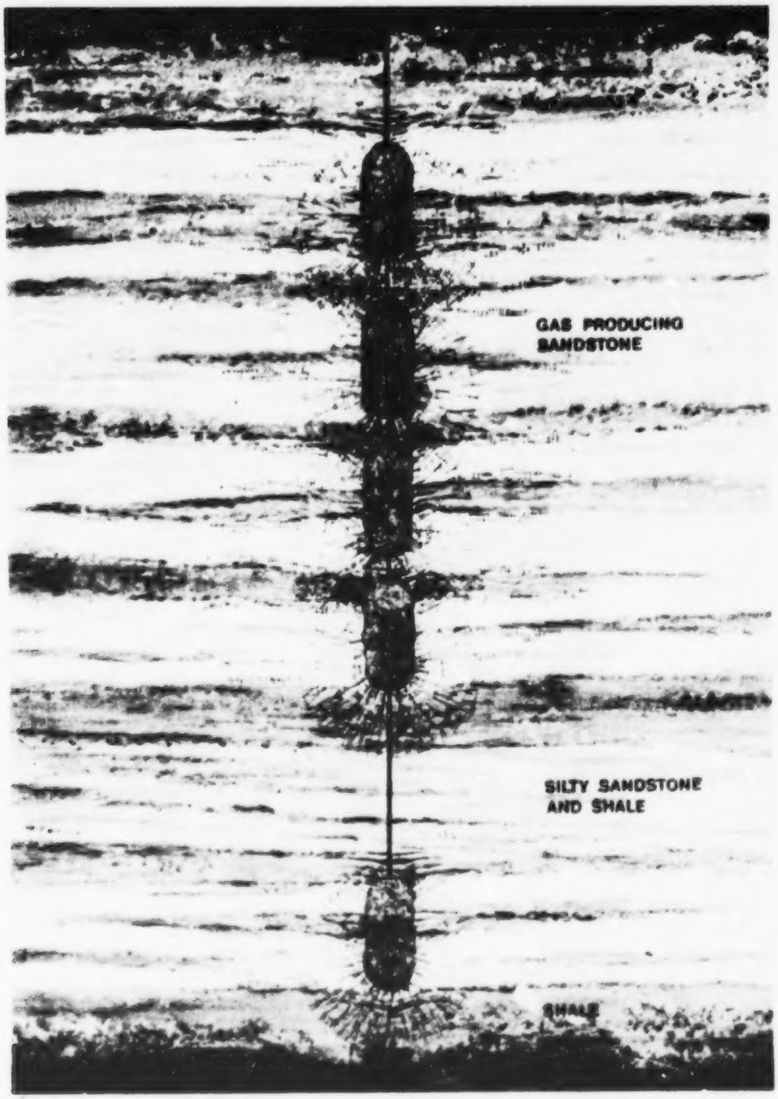
Stimulation of Gas and Oil Reservoirs: Many fields exist in which the large quantities of gas and oil present cannot be recovered economically because of the low permeability of the reservoir rock. Plowshare is developing techniques to fracture the reservoir rock with nuclear explosions and thus, increase the permeability.

When a nuclear explosive is detonated underground in a completely contained explosion, it creates a cavity and fractures the surrounding rock out to relatively large distances. Figure 1 shows a series of five explosives emplaced in the same hole to increase the premeability of the rock and speed the flow of gas or oil. The principle has been successfully demonstrated with one explosive in the Gasbuggy Event in northwestern New Mexico (December 1967, 29 kt) and one in the Rulison Event in western Colorado (September 1969, 40 kt). In addition, the U.S.S.R. has conducted one gas and two oil-stimulation experiments using a total of eight nuclear explosives.

In this application the main radioactive contaminants of the gas produced are the volatile radionuclides (e.g., tritium and ^{85}Kr) that mix with the natural gas. Tritium is especially important, as a small fraction of it replaces the normal hydrogen in natural gas during the explosion and, thus, is practically impossible to remove. Some tritium remains trapped in the vicinity of the detonation; for example, in Gasbuggy and Rulison, only 5-10 percent of the total residual tritium was found in the gas. The nonvolatile nuclei also are trapped underground. Thus, a fission explosive is preferred since it makes considerably less tritium than does a fusion explosive. In this regard, we find that in addition to volatile fission products, tritium from the ternary fission by fission-spectrum neutrons must be considered. As previously stated, tritium is also produced by neutrons reacting with lithium in the rock.

Figure 1

Stimulation of Natural-Gas Production Extensive Fracturing of Reservoir Rock With Nuclear Explosives

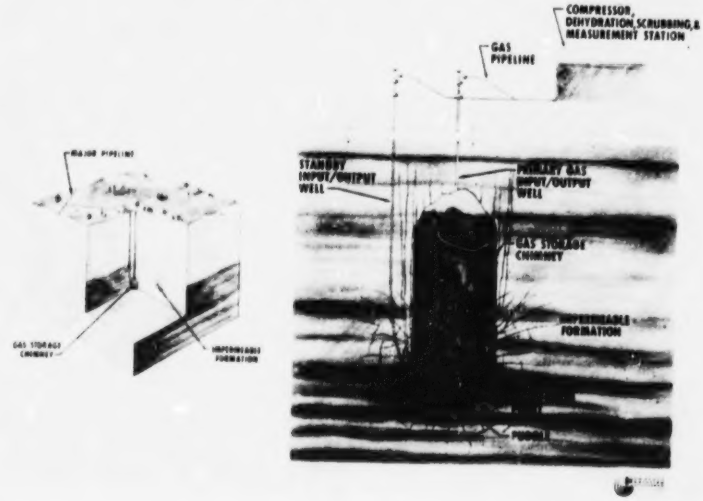


The next planned gas stimulation experiment is called Rio Blanco. This experiment, in the northern Piceance Basin of Rio Blanco County, Colorado, should demonostrate the effectiveness of gas stimulation by tandem emplacement of three explosives of <8 inches in diameter placed between depths of 5,800 and 6,800 feet. These three explosives, detonated simultaneously at yields of 30 kt each, would be used so that the total maximum credible yield would be less than 100 kt. This experiment is tentatively scheduled for late summer of 1973. In the Rio Blanco experiment, the three 30 kt explosives are expected to leave behind <3 kCi of tritium or on the average <1 kCi of tritium for each 30 kt explosive.

Gas Storage: There has been a tremendous growth in the use of natural gas as an energy source. Its advantage over other energy sources, from an air pollution standpoint, should cause this growth rate to continue, provided that proven gas reserves are adequate. Fluctuations in demand for gas from time to time cause a major problem for the gas-distribution industry. In order to avoid having to build large pipeline facilities adequate to supply peak demand, a capability for the storage of large volumes of gas is required. Figure 2 illustrates a concept to satisfy this requirement. A contained underground nuclear explosion could be used to create a cavity or chimney in a very tight, unfractured, impermeable formation such as salt or shale at a location as close to the market as is seismically allowable. Here we see an underground void space that could be used to store gas after the radioactivity has been removed, immobilized, or allowed to decay to acceptable levels.

Figure 2

Gas-Storage Reservoirs Could be Created by Plowshare Underground Nuclear Explosions.

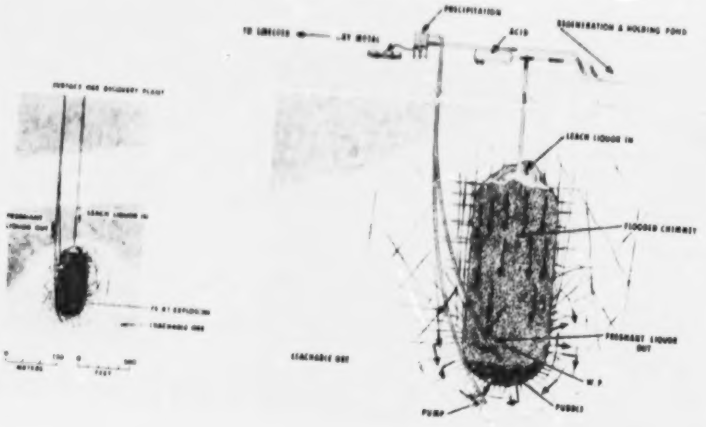


Calculations show that the radionuclides that might cause the greatest problem in this application are tritium, the fission product ^{85}Kr, and, where the detonation takes place in salt, the ^{35}S produced by the neutron reaction on ^{35}Cl. In salt, the lithium concentration is normally extremely small, so most of the tritium will come from the nuclear explosive. The 87-day half-life of ^{35}S could either be reduced by neutron shielding or merely by waiting for it to decay. Whether a fission or fusion explosive would be used for creating a gas-storage reservoir would depend partly on the relative radiological safety impact of tritium and ^{85}Kr.

Mineral Leaching: Throughout the world a large number of copper-ore deposits exist where the copper is too deep, too small in absolute quantity, or distributed too diffusely to be economically recoverable by conventional mining techniques. For those deposits that are deep enough, a contained underground nuclear explosion could be used to create a chimney and fracture zone in the ore deposit. This is shown in Figure 3 for the case of oxide ores. An acid solution could then be introduced at the top of the chimney and percolated down through the broken and fractured ore, leaching the copper or other minerals of interest. The leach solution with the dissolved copper would be pumped from the bottom of the chimney to the surface, where conventional separation facilities would remove the copper and return the acid to the top of the chimney for another cycle. A leaching method applicable to sulfide ores involves pumping oxygen down to the bottom of the chimney after the chimney has been filled with water. The oxygen would oxidize the sulfur in the copper-bearing sulfide ore to form dilute sulfuric acid. For this application, a fusion explosive may be more desirable, if specific fission products that leach out with the copper are more of a problem than is the tritium from the fusion explosive. It is not entirely clear at this time, however, whether either the fission products or the tritium will present a problem, and hence both must be given consideration.

Figure 3

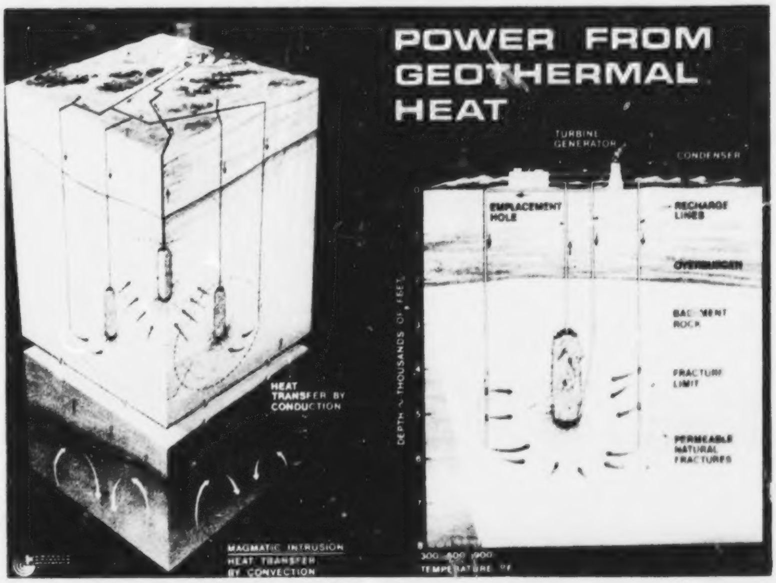
Plowshare Application of an Underground Nuclear Explosion to Crush Ore for *in situ* Leaching



Geothermal Power: In many geothermal areas of the world, a natural system of underground fractures interacts with a water-circulation system and heat source to form an underground reservoir of steam or hot water. Steam is obtained by drilling into such reservoirs. In other geothermal areas where steam production is limited because of inadequate or plugged natural fracture systems, an underground nuclear explosion could be used to create a chimney and an associated fracture system (Figure 4) and to augment existing fractures. Heat might also be extracted directly from the heat source itself when no reservoir or water-circulation system is present. In this case, an underground nuclear explosion would be used to fracture the hot source rock, water would be introduced into this chimney-fracture system, and the resulting superheated steam would be removed for the generation of electrical power. Here again, the amounts of volatile radioactive species (primarily fission products plus tritium and the Argon 37 that is formed by neutrons reacting with calcium in the rock) that might contaminate the steam must be considered.

Figure 4

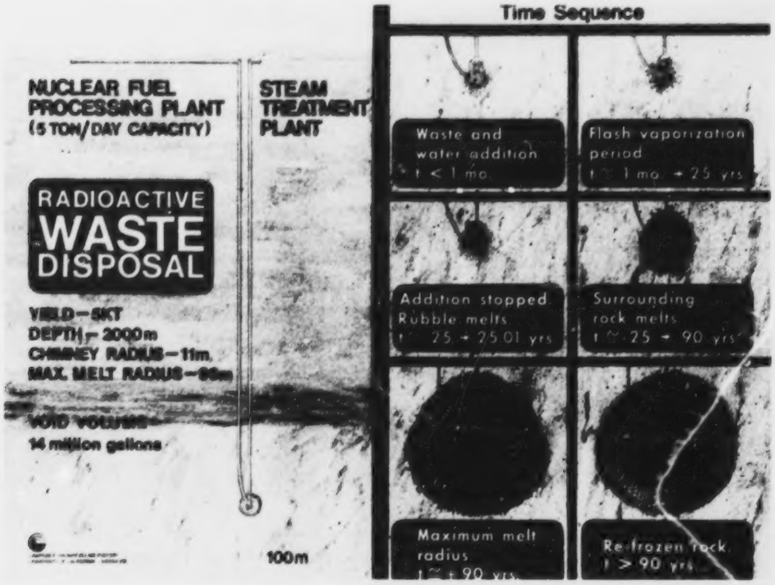
Plowshare Application of Nuclear Explosives to Fracture Rock in Geothermal Areas for Development of Geothermal Power



Waste Disposal: Another underground application under consideration is radioactive waste disposal, wherein the radioactive waste from fuel-reprocessing plants would be stored in underground chimneys created by contained nuclear explosions. This concept is shown in Figure 5 [2]. One or more chimneys would be created at the site of the fuel-reprocessing plant. Radioactive waste would be injected into the chimney and allowed to self-boil. When it had served its purpose, the chimney would be boiled dry and cemented shut. The heat generated by the waste would then melt the rock, which would dissolve the waste material. When the molten material cooled, the radioactivity would be permanently and insolubly incorporated into the rock. The amount of tritium produced by the explosive in this application is relatively unimportant, because the amount of radioactivity injected into the chimney would be much greater than the amount produced by the nuclear explosive. Most of the tritium is expected to exchange with the water pumped down the chimney and any molecular tritium could be catalytically removed. Since this method involves total recycling of water, under normal operating conditions essentially no tritium would be released to the environment from either fuel reprocessing or storage operations.

Figure 5

Plowshare Proposal for Radioactive Waste Disposal


Time Sequence
NUCLEAR FUEL PROCESSING PLANT (5 TON/DAY CAPACITY)
STEAM TREATMENT PLANT
RADIOACTIVE WASTE DISPOSAL
YIELD—5KT
DEPTH—2000m
CHIMNEY RADIUS—11m
MAX. MELT RADIUS—90m
VOID VOLUME—
14 million gallons
100m
Waste and water addition t < 1 mo.
Flash vaporization period t ≃ 1 mo → 25 yrs
Addition stopped. Rubble melts t ≃ 25 → 25.01 yrs
Surrounding rock melts t ≃ 25 → 90 yrs
Maximum melt radius t ≃ 90 yrs
Re-frozen rock t > 90 yrs

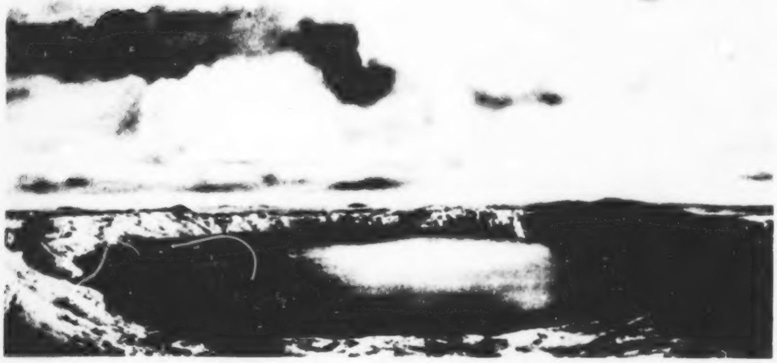
Excavation: The chief Plowshare nuclear excavation proposals have been for the construction of a sea-level canal near the present Panama Canal, the construction of harbors — especially in Alaska and

Australia — and the use of nuclear craters for developing water reservoirs. In these concepts we have looked at the tritium problem in detail.

The final report to the President by the Interocean Canal Study Commission [3] states that, "Although we are confident that someday nuclear explosives will be used in a wide variety of massive earth-moving projects, no current decision on the United States canal policy should be made in the expection that nuclear excavation technology will be available for canal construction." Recommendation 7A indicates that, "the United States [should] pursue development of the nuclear explosive technology." Thus, although there is little research currently being carried out by the U.S. in the area of nuclear excavation, this technology may find future application in remote regions of the world. It should be pointed out that the U.S.S.R. has built a reservoir — which is now in use — with a nuclear explosive. This reservoir is shown in Figure 6. A movie released by the U.S.S.R. and shown at the Symposium, shows a man diving and swimming in this reservoir. The yield of the explosive used here was a little larger than 100 kt and the explosion created a crater having a maximum depth of 100 m and a mean diameter, at the original earth surface, of over 400 m giving a crater volume of 6,300,000 m^3.

Figure 6

Reservoir Excavated in U.S.S.R. With a Nuclear Explosion



Excavation applications require explosives that produce relatively small amounts of fission products and induced activities. Thus, a thermonuclear explosive with a very small yield from the fission trigger must be used, and residual tritium would be one of the main concerns. Extensive neutron shielding is necessary, and materials in and around the explosive that can produce radioactive products that are particularly objectionable from a biological viewpoint are replaced by materials that give fewer undesirable products.

Calculational Method

Although similar methods are used for excavation and underground engineering applications, this discussion pertains primarily to gas stimulation. Since the tritium production by a nuclear detonation has become such an important consideration, considerable effort has been made in developing the neutron-activation calculational methods [4]. To accurately calculate the activation products, a number of parameters must be known: these include the neutron flux level within the materials of concern, the temperature of the materials at the time of neutron capture (since energy from the explosive will heat surrounding materials to very high temperatures), changes in the materials' configuration or geometry resulting from hydrodynamic effects, and both the neutron scattering and various capture cross sections of the materials, including all the constituents of the rock.

The neutron flux level within a certain material is calculated using both static (fixed geometry) and dynamic (changing geometry) multi energy-group Monte Carlo neutronic computer codes. By geometry we mean the boundaries, densities, and locations of the various materials vaporized in the explosion. It is necessary to employ the hydrodynamic code initially, to determine the number of neutrons that escape the explosive as well as the temperature and geometry or configuration of the material and rock surrounding the explosive. Once these values are known, a static Monte Carlo code is used to transport the neutrons that do escape the explosive throughout the surrounding environment.

Once the neutron flux has been determined for a certain region, the radioactive products can then be calculated; the computer code used is ACTIVE. ACTIVE will compute the products of all reactions contained in its cross section library [5] for a given material in a zone by multiplying the appropriate cross sections by the flux. The ACTIVE cross section library contains approximately 800 different reactions. Zonal products are then added to obtain the total activation products at time zero. Since the total activation takes place as a result of a single burst of neutrons over a very short period of time and the flux levels outside of the explosive are relatively low, decay during activation and second- or multiple-order reactions are not calculated by ACTIVE. Delayed fission neutrons are also not considered, because of their relatively small abundance of less than one percent. The time zero products are then decayed according to their radioactive half-lives to obtain activities as a function of time.

This calculational method is also used to study the effects of rock composition, especially lithium and boron, on the tritium production from the rock and to determine the effectiveness of various shielding materials.

It should be mentioned that the accuracy of this method depends strongly upon the reliability of the different cross sections employed by the computer codes [6]. For some materials, cross sections have never been measured, and these must either be calculated by using nuclear models or extrapolated from other known cross sections. In making the transfer from the dynamic to the static case, certain assumptions must be made regarding such parameters as the nuclear temperature and geometric configuration. These also affect the accuracy of the calculations. It is difficult to estimate the cumulative effects of these uncertainties on the calculations; certainly the effects will be different for each specific case. The effect of varying one of these parameters (the temperature of the rock) will be discussed later in the paper.

Tritium Reduction

To minimize the amount of tritium produced for any particular application, a fission explosive would be used. This would then establish the lower limit of tritium production as that produced by ternary fission, which is equal to about 1 Ci(0.1 mg)/kt. As has been mentioned previously, there are other sources of tritium external to the explosive, the largest usually being from neutron capture by lithium in the rock. For rock containing approximately 30 ppm (by weight) of Li, which is found in some natural-gas-bearing media, 1 to 3 percent of the neutrons escaping into the rock will produce tritium. The amount of tritium that is formed will be dependent on the amount of lithium and other absorbers present in the rock and on the temperature of the rock at the time of neutron capture. We have calculated that temperatures in the range of 1 eV to 1 keV (where 1 eV = 12,000°K) are typical for the rock surrounding the explosives. Within this range, the tritium production rate changes by a factor of approximately seven. Since neutron capture takes place throughout this entire range, the total production varies by only about a factor of three.

We have previously determined [7] that borated polyethylene is an attractive external shielding material for Plowshare applications, considering both its cost and its neutronic properties. Employing a shield of this type will reduce the amount of tritium formed in the rock, although some tritium is formed from the boron in the shield. An evaluation as to the effect on the total tritium produced when employing a shield of this type would have to be made for each particular application.

Conclusions

The total residual tritium from the 29-kt Gasbuggy Event was 40 kCi, and the total residual tritium from the 40-kt Rulison Event was 10 kCi. This is similar to the amount of tritium produced by a 1,000 MW/year reactor [8]. Through the design of the LLL Diamond family of underground-engineering explosives, about 1 kCi of total residual tritium were expected from the 80-kt Plowshare Miniata Event fired at the Nevada Test Site this July. Miniata was a test of one of the members of the Diamond family of explosives developed especially for use in gas stimulation projects. The Diamond explosive is expected to produce less than 2 kCi of residual tritium at yields of 100 kt in a gas stimulation application. It should be understood that these numbers are based on a rock composition that has been observed as being representative of natural-gas-bearing media. A large change in the amount of lithium or other neutron-absorbing materials in the rock could affect the tritium production accordingly.

The large decrease in tritium production from approximately 1.4 kCi/kt for the Gasbuggy Event to 12 Ci/kt for the Miniata Event, a reduction of over a factor of 100, is indicative of the progress that has been made by the Plowshare Program toward the goal of minimizing the tritium while developing an explosive system that is attractive for industrial application.

ACKNOWLEDGMENT

This report is based on work performed under the auspices of the U.S. Atomic Energy Commission.

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PRODUCTION OF TRITIUM BY NUCLEAR WEAPONS

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The tritium production by a nuclear weapon is obviously closely related to its specific design and to the environment in which it is detonated. This paper discusses mainly the tritium that results from nuclear reactions in thermonuclear fuel. The production of tritium by interaction of nuclear reaction products with the immediate environment, i.e., tritium that is not a result of interactions within the fuel, is covered in some detail by Green and Lessler [1].

We can consider two limiting classes of nuclear weapons: those in which all the energy is produced by the fission process and those in which all the enrgy is produced by fusion reactions. Although the latter case is not within the current technology, it is convenient to use it as a model for discussing tritium production.

In order to establish a "relative significance index" for the tritium produced by nuclear weapons, the amount of tritium in our environment resulting from natural cosmic processes will be used as a standard.

Fission Weapons

The tritium from an all-fission weapon can be seen to be relatively unimportant by comparison with the tritium in nature. The production of tritium in nature has been estimated to give rise to a steady-state tritium inventory of about 100 MCi [2].

The yield of tritium in the fission process is approximately $10^{-2}\%$ [3]. The production of 1 Mt of energy by fission requires 1.4×10^{26} fissions and produces about 700 Ci (0.07 g) of tritium. Thus, if tritium were our only concern, the steady-state detonation of about 20 Mt per day would produce a tritium burden just equal to that occurring in nature. Therefore, it is reasonable to consider the direct production of tritium by fission weapons as an unimportant contribution to the world inventory.

Thermonuclear Weapons

Let us now consider thermonuclear weapons. For a device with $^{6}Li^{2}H$ fuel, one could write:

1) $^{6}Li + n \rightarrow {}^{4}He + {}^{3}H$ 110 kt/kmol

2) $^{3}H + {}^{2}H \rightarrow {}^{4}He + n$ 405 kt/kmol

1) + 2) = 3) $^{6}Li^{2}H \rightarrow 2\,{}^{4}He$ 515 kt/kmol

Reaction 3), the sum of 1) and 2), would appear to result in the conversion of $^{6}Li^{2}H$ to ^{4}He with no production of tritium, a most desirable attribute.

The actual situation with respect to thermonuclear weapons is far more complicated, however. Other nuclear reactions between the species initially present in the fuel or those produced during thermonuclear burn must be considered. Some examples of possible interactions are:

4) $^{2}H + {}^{2}H \rightarrow n + {}^{3}He$ +85 kt/kmol

5) $^{2}H + {}^{2}H \rightarrow {}^{1}H + {}^{3}H$ +93 kt/kmol

6) $^{2}H + {}^{3}He \rightarrow {}^{4}He + {}^{1}H$ +423 kt/kmol

7) $n + {}^{3}He \rightarrow {}^{1}H + {}^{3}H$ +18 kt/kmol

8) $n + {}^{2}H \rightarrow 2n + {}^{1}H$ -51 kt/kmol

9) $^{2}H + {}^{6}Li \rightarrow {}^{4}He + {}^{4}He$ +515 kt/kmol

The rates of these and other possible reactions are strongly dependent on the temperature and concentrations of the reactants and products; the competition between them is intimately related to the detailed weapon design. The relationship between total energy released and tritium produced is, thus, not a simple one.

In a more general form, one could rewrite Reaction 3) as:

$$10)\quad a_1\,{}^{6}\mathrm{Li} + a_2\,{}^{2}\mathrm{H} \rightarrow a_3\,{}^{4}\mathrm{He} + a_4\,{}^{3}\mathrm{He} + a_5\,{}^{1}\mathrm{H} + a_6\mathrm{n} + a_7\,{}^{3}\mathrm{H},$$

where the parameters $a_1 \ldots a_7$ are dependent on the weapon design, and the net mix of the reactions almost certainly does not result in the simplicity of Eq. 3.

The yield of tritium from thermonuclear explosives has been reported to be between 0.7 and 5 kg per megaton of yield [4]. The fairly wide range in reported yields reflects the possible variations in design and performance of individual explosives. For purposes of discussion, 2 kg (~20 MCi) of tritium per megaton will be used as an average value.

The first thermonuclear explosions occurred in 1952. Since that time, tests by the nuclear powers have included both fission and fusion weapons. With the exception of cessation of testing during the voluntary moratorium from October 30, 1958 until September 1, 1962, there have been some tests during each calendar year.

The availability of tritium in the thermonuclear tests is, of course, quite different from surface and above-ground detonations than from underground detonations. In reviewing the contributions of nuclear weapons explosives to the world tritium inventory, it is advantageous to treat the underground tests separately from the others.

Atmospheric Testing

Izrael and Stukin [5] have summarized the total yields of all nuclear-weapon tests from 1945 through 1962. They report the following data:

	Total yield (Mt)	Fission yield (Mt)	Fusion yield (Mt)
1945-1958	170	92	78
1959-1962	510	190	320
Total	680	282	398

The total tritium produced in these tests is approximately 8 GCi. No thermonuclear explosions are reported before 1952, and there were no atmospheric tests during 1959, 1960, or 1961. The amount of tritium currently in the environment from atmospheric testing can be estimated by assuming that injections have occurred as two pulses, one from 78 Mt of fusion in 1955 and one from 320 Mt of fusion in 1962. This would give an inventory at this time of 4,500 MCi, or about 45 times that due to the natural processes.

In addition to the tritium from the tests cited above, there have been some thermonuclear tests by countries that have not signed the Limited Test Ban Treaty. The total amount of tritium from these tests appears to be small, however.

Underground Testing

Since mid-1962, the U.S. and the U.S.S.R. have conducted all their nuclear weapons tests underground. The immediate availability of the tritium to the biosphere is obviously greatly reduced under these conditions. The behavior of tritium in ground water is discussed by F. W. Stead [6], and the behavior in soil by G. L. Stewart [7]. Following comments relate to the possible direct contributions of tritium to the atmosphere from underground weapons tests.

In order to evaluate the effect of underground tests of thermonuclear devices on the "available" world tritium inventory, we must attempt to estimate the fraction of the tritium produced that might be released to the atmosphere. In this discussion "available" is used to mean tritium injected into the atmosphere or deposited on the surface of the ground.

The availability of the tritium will depend on its chemical form and on whether or not the test was completely contained (i.e., no detectable amount of radioactivity released through cracks, crevices, stemming failures, etc.).

Taylor has estimated the distribution between H_2O and H_2 of the tritium produced in an underground nuclear explosion under various test conditions [8]. The most important parameters are the amounts of iron and water available for chemical reactions. His results are shown in Figure 1.

A reasonable upper limit for the fraction of the tritium that is in elemental form appears to be one percent. In any venting process we can assume that the fraction of the H_2 released will be the same as the fraction of the rare gases. (In actuality this should be an upper limit.) In a dynamic vent where there is a direct connection between the source and the surface, we can assume that the tritiated water will also be released to the same extent as the rare gases. Again this obviously is an upper limit. In any venting that involves passage of the vented materials through earth, cables, or tortuous paths, we would expect the tritium present as water to be effectively scavenged from the release.

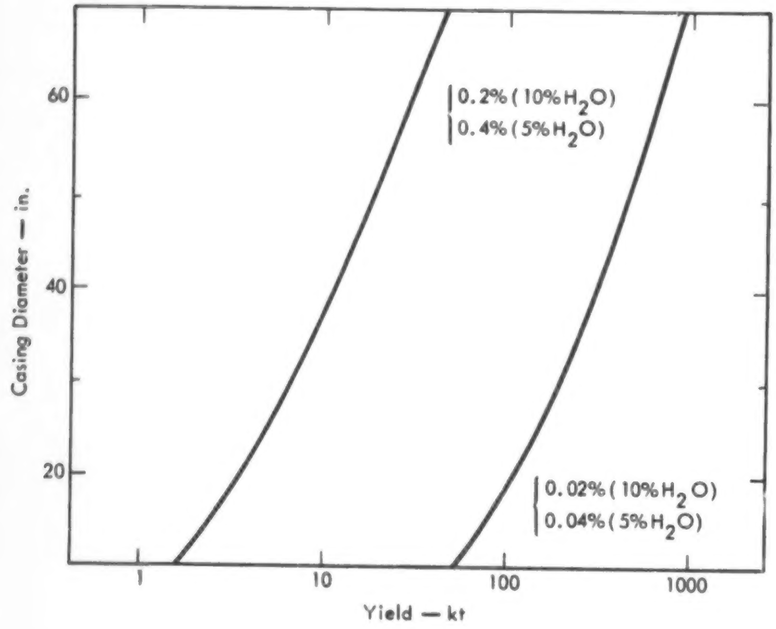
The U.S. has announced 230 underground tests since the signing of the Limited Test Ban Treaty [9]. Of these, five have been cratering experiments which, although detonated underground, do release a larger fraction of the radioactivity produced to the atmosphere than do the contained tests. Of the remaining 225, 17 have vented radioactivity which was detected off the test site. Of the 17 tests that have vented radioactivity, 15 are reported as having had a "low" yield ("low" in this context is defined as <20 kt), one as "low-intermediate" (i.e., 20-200 kt), and the other as 1.2 kt. Assuming that the maximum yield in each category applied, the total yield of these 17 events is approximately 500 kt. These events were fairly uniformly distributed in time during the eight-year period reported. Thus, the average annual yield-rate, based on the previous assumptions, would be about 60 kt/a.

Assuming that all of these devices were pure thermonuclear, then the total tritium produced per year by them would be 1.2 MCi.

The Radiological Effects Section of the AEC, Nevada Operations

Figure 1

Postshot H_2/H_2O distribution in cased holes. The lines show the relation among yield, casing diameter, and water content of the rock, with the percentages of total tritium as ^{1}HT [8].

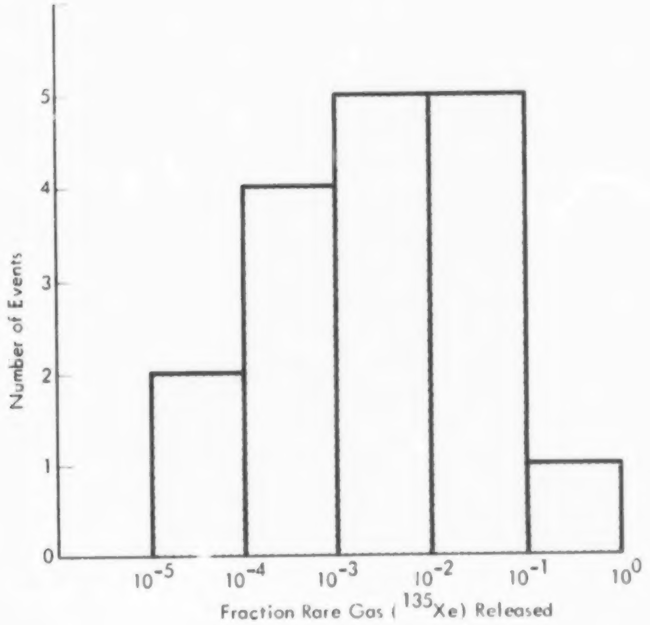


Office, has summarized the number of curies released by each of the 17 tests mentioned above [10]. Examination of the data reveals that if all the activity released were assumed to be ^{135}Xe, and if the fission yield of the "low" yield devices is taken as 10 kt and that of the "low-intermediate" as 50 kt, then the release fractions for the ^{135}Xe range from 10^{-5} to 1. (A plot of the number vs. release fraction is shown in Figure 2.) The median release fraction is in the range 10^{-3} to 10^{-2}.

If we combine the fraction of tritium expected to be in the form of hydrogen gas with the above estimates on fractional release, we obtain, for the contribution to the atmospheric tritium inventory from these test releases, a range of from 1.2 X 10^{-7} MCi to 1.2 X 10^{-2} MCi per year, with a median value of about 1.2 X 10^{-4} MCi. Even the upper limit is small in comparison with the "natural" inventory of 100 MCi.

Figure 2

Rare Gas Release in Underground Tests



Conclusions

The effects of nuclear weapons tests on the current world tritium inventory can be summarized as follows:

1. The contribution from fission weapons is negligible, whether they were tested in the atmosphere or underground.
2. The contribution from the thermonuclear tests that were conducted in the atmosphere is large. The present invertory is approximately 45 times the natural background as a result of weapons testing and, even in the absence of further atmospheric tests, will be a significant (>10%) perturbation on the natural background for about 100 years.

3. The underground testing of thermonuclear weapons has not contributed significantly to the atmospheric burden, even when the containment has been imperfect.

ACKNOWLEDGMENT

This report is based on work performed under the auspices of the U.S. Atomic Energy Commission.

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CHAPTER THREE
Detection and Measurement

THE CALIBRATION OF THE NATIONAL BUREAU OF STANDARDS' TRITIUM STANDARDS BY MICROCALORIMETRY AND GAS COUNTING

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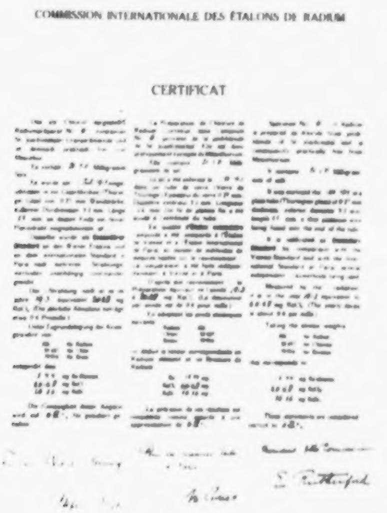
Washington, DC

By way of introduction, Figures 1 and 2 show the certificates of the first radioactivity standards acquired by the National Bureau of Standards, those of the 1913 and 1936 radium standards from the International Radium Standards Commission. These standards are defined standards based on a mass of radium salt, similar in this respect to the international standards of mass, length and time. Over the years, however, the activity of a gram of radium was measured by a number of investigators and it was found to lie between 3.6×10^{10} and 3.7×10^{10} disintegrations per second per gram of radium.

Figure 1

Certificate of the International Radium Standards Commission for the first United States Primary Radium Standard, Vienna No. 6 and International Commission No. IV, which was brought to America in 1913 by Mme. Marie Curie. It is interesting to note the somewhat large difference in the mass of the radium chloride as determined by weighing and gamma-ray measurement.

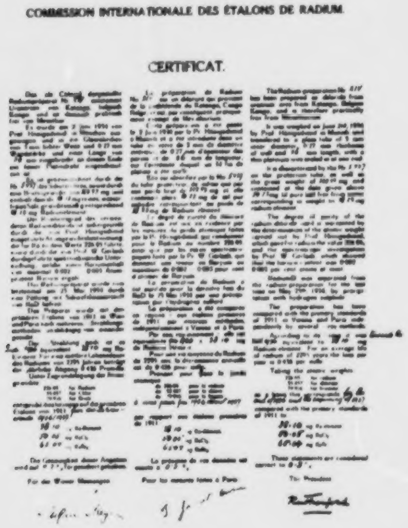


COMMISSION INTERNATIONALE DES ÉTALONS DE RADIUM

CERTIFICAT

Figure 2

Certificate of the International Radium Standards Commission for United States Primary Radium Standard, No. XIV. This standard which is Honigschmid standard No. 5437, is actually about 36.7 mm in length. According to Stefan Meyer (1945), the Paris and Vienna gamma-ray measurements of the amount of radium chloride in this standard were respectively 50.03 and 50.08 mg.



COMMISSION INTERNATIONALE DES ÉTALONS DE RADIUM.

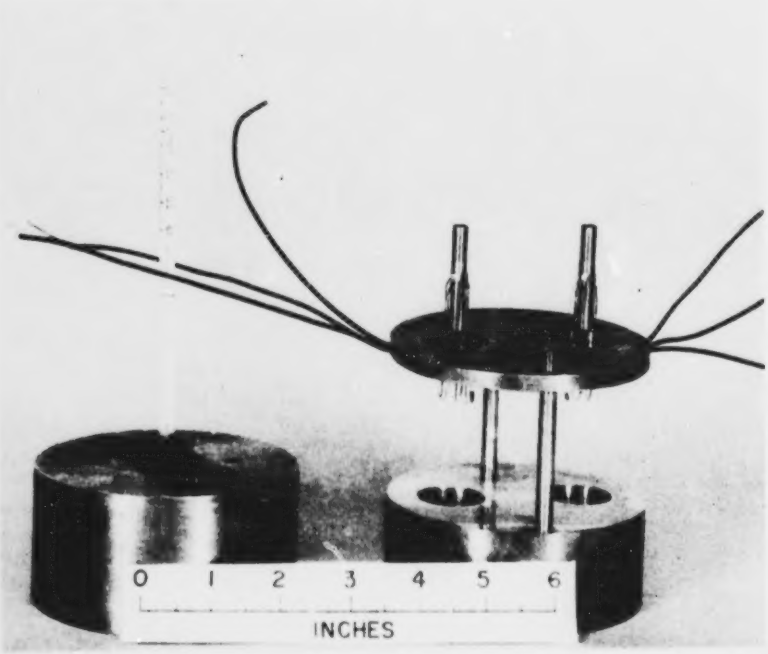
CERTIFICAT.

Under the impact of the availability of artifically produced radionuclides after World War II, the VIth International Congress of Radiology, meeting in London in 1950, defined the curie as the quantity of any radioactive nuclide in which the number of disintegrations per second is 3.7 X 10^{10}. Previously the curie had simply been the quantity of radon in equilibrium with 1 g of radium.

In 1953, a Peltier-twin-cup microcalorimeter was constructed for the measurement of a number of international radium standards prepared by Hönigschmid in 1934, and of which two comprised our 1936 radium standards [1-3]. This microcalorimeter, shown in Figure 3, was subsequently used, in 1954, to prepare the first National Bureau of Standards' tritium standards. There never has been any formal publication of this work, but it has been briefly described in two reviews [4,5].

Figure 3

The radiation balance or twin-cup Peltier-effect microcalorimeter that was constructed for the comparison of a number of national Honigschmid radium standards. The cups of this radiation balance were of gold and it was unclosed in a temperature-attenuating enclosure to give a precision in the comparison of the national standards of about ±0.1 percent. The scale sensitivity, which limits the precision of the lowest rate of energy dissipation measurement, is about 3 mm/μW.



In these measurements a sample of approximately 4.8 Ci of tritiated water was sealed in a small quartz tube in an atmosphere of hydrogen at a pressure of about 2 cm Hg, in order to inhibit the formation, by irradiation of the water, of H and OH radicals. The rate of energy emission from this sample was measured over a period of four months in order to be assured that no transient transfer of chemical energy might be taking place. The results obtained are shown in Table 1.

Table 1

Rate of Energy Emission from Quartz-Encapsulated Sample of Tritiated Water As Measured in NBS Radiation Balance

Date	Rate of energy emission (μW)	Activity (c)	Activity (c) corrected to August 20, 1954
August 20, 1954	163.5_9	4.85_0	4.85_0
	$[118.3_8]$	$[3.51_0]$	$[3.51_0]$*
August 25, 1954	162.0_9	4.80_6	4.81_0
	$[117.8_7]$	$[3.49_5]$	$[3.49_8]$*
August 30, 1954	161.3_1	4.78_3	4.79_0
September 8, 1954	162.6_8	4.82_3	4.83_7
October 5, 1954	162.8_6	4.82_9	4.86_3
November 2, 1954	160.1_9	4.74_9	4.80_4
November 30, 1954	158.3_6	4.69_5	4.77_0
December 28, 1954	159.3_7	4.72_5	4.82_1
May 10 and 11, 1955	155.6	4.61_3	4.80_5

*The quartz enclosure of this sample cracked on August 26, 1954.
Average: 4.81_7 c on August 20, 1954 (standard error = 0.2 percent).

After these measurements the quartz ampoule was crushed under 1 kg of water in a specially constructed stainless steel container, and the master solution so obtained was diluted to give standards of activity of around 10^4 and 10^5 s^{-1} ml^{-1}, which were flame-sealed into glass ampoules for distribution.

The exact rate of disintegration per milliliter was obtained from the rate of energy emission per milliliter and the assumed mean energy of 5.69 keV per disintegration for hydrogen-3 (with a probable error of 0.04 keV) [6,7].

Even though the measurement of the rate of energy emission was direct, these tritium standards were, however, only relative standards that were based ultimately on the disintegration rate measured by Jenks, Ghormley, and Sweeton, who determined it by the rate of production of helium-3. It was, therefore, imperative that the NBS hydrogen-3 standards should be measured independently. This was subsequently carried out in 1961 by the method of gas counting, using the NBS length-compensated gas counters which had previously been used for the preparation of the NBS carbon-14 activity standards [8] and for the measurement of the half-life of carbon-14 [9]. These counters, which were damaged in moving to our new laboratories, have been reconstructed and recalibrated and are shown in Figures 4 and 5 in their original and present condition. The principle of their operation is illustrated in Figures 6 through 10.

Figure 4

The National Bureau of Standards length-compensated gas counters for the counting of low-energy beta emitters and electron-capturing nuclides assembled within a steel shield and with their auxiliary gas-handling equipment.

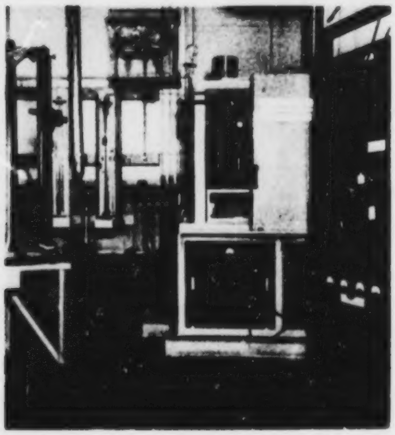


Figure 5

NBS length-compensated gas counters in new vacuum housing

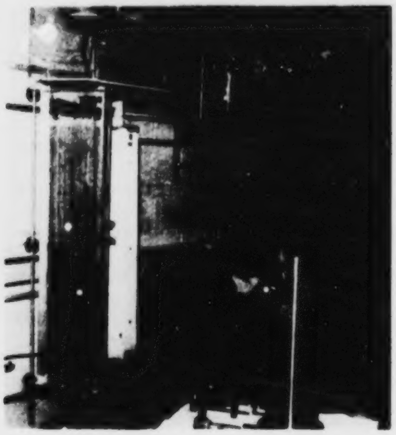


Figure 6

A set of three stainless-steel compensated counters with glass and stainless-steel ends. The glass insulators are cemented into stainless-steel rings which are in turn screwed on to the stainless-steel cathodes by means of carefully cut micrometer threads. The ends were mounted onto the cathodes using a traveling microscope to insure that the distance from the end of each cathode to the stainless-steel disk supporting the tungsten anode wire was the same in all of the counters. The cathode lengths of the stainless-steel counters are 3.7586, 5.7410, and 11.7673 in. The counters have recently been reconstructed with teflon and stainless-steel insulationg ends.

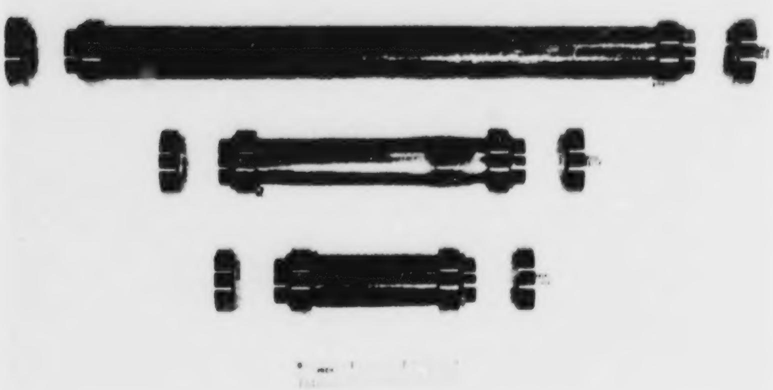


Figure 7

Schematic distribution of electric field in internal gas counters.

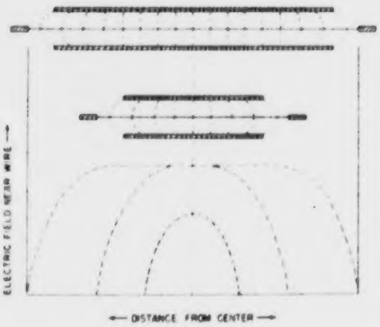


Figure 8

Internal gas counter plateau

COPPER COMPENSATED GAS COUNTERS

PRESSURE 750.92 X 10³ DYNES/CM²

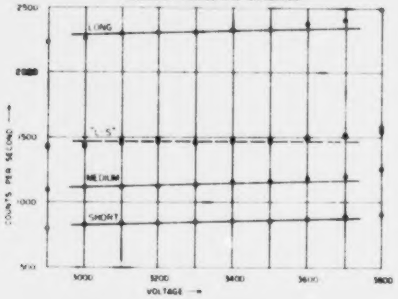


Figure 9

Internal gas counter plateau

STAINLESS STEEL COMPENSATED GAS COUNTERS

PRESSURE 750.92 X 10³ DYNES/CM²

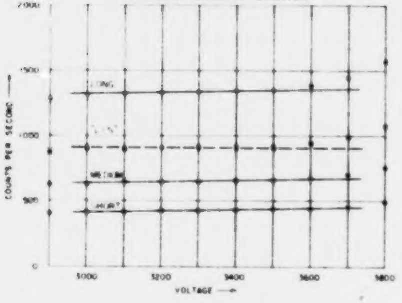


Figure 10

Differential discriminator readings

LONG COPPER COUNTER MINUS SHORT COPPER COUNTER

(MESSAGE REGISTERS x 256 PER MINUTE)

	10	20	30	40	50	60	VOLTS (DISCR)
3300	649 240 409	630 234 396	615 231 384	615 230 385	602 223 379	603 220 383	
3400	662 248 414	648 241 407	645 240 405	639 236 403	636 233 403	628 233 395	
3500	697 244 453	658 240 418	654 246 408	650 239 411	645 239 406	634 241 393	
3600	778 245 533	661 245 416	654 244 410	652 238 414	649 239 410	646 248 398	
3700	905 257 648	661 252 409	656 247 409	651 252 399	663 247 416	650 250 400	
VOLTS (CATHODE)							

For the purpose of counting, the selected tritium standards had to be reduced to hydrogen by passing the water vapor over zinc at 350°C and the resulting hydrogen was then quantitatively mixed with methane for gas counting. The apparatus used for the preparation of the hydrogen is shown in Figure 11. Because of the possibility of isotopic separation, which is illustrated in Figure 12, it was necessary to carry the chemical reduction to completion. The internal gas-counting measurements were carried out in the proportional region and errors due to wall effects and to loss of small pulses were investigated by measuring the count rates as a function of pressure and discrimination level. The experiments have been fully described [10] and the final results are summarized in the certificate shown in Figure 13. The value of activity obtained by gas counting in 1961 is 1.96% lower than that obtained microcalorimetrically in 1954. In view of the uncertainty in mean energy, this result is not unsatisfactory.

Figure 11

Schematic drawing of the apparatus used for preparing the hydrogen samples for gas counting.

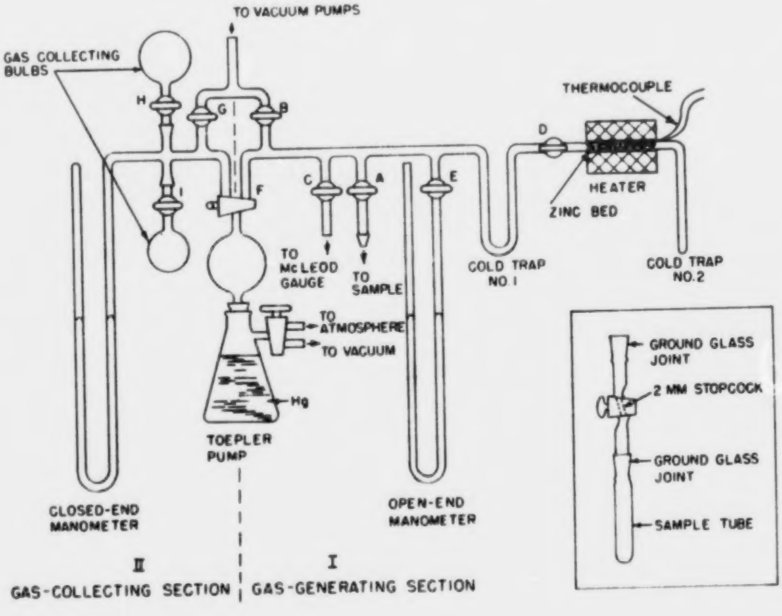


Figure 12

Activity of each sample expressed in mV/sec/g mM (as measured in the vibrating-reed electrometer) as a function of the fraction of the total g mM of water decomposed. The crosses and circles represent the results of the first and second runs, respectively.

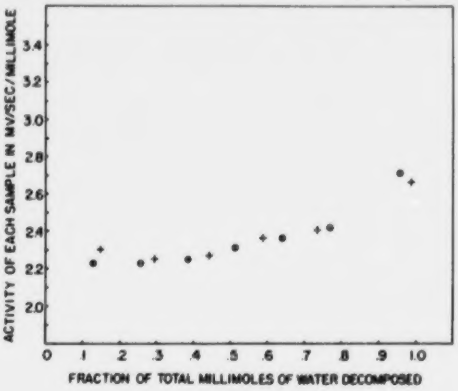
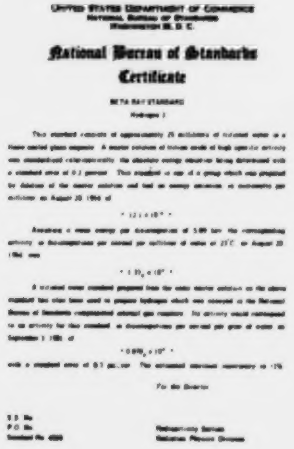


Figure 13

New certificate issued for NBS radioactivity standard sample 4926 giving both the original microcalorimetric and more recent gas-counting results. The use of the gas-counting results is now, however, to be preferred. The mean energy per disintegration is a calorimetrically determined value [6,7] so that no correction for re-arrangement of the extra-nuclear electrons is needed.



National Bureau of Standards

Certificate

BETA RAY STANDARD

Subsequently, these standards were measured by the Euratom Geel Laboratory [11]. The agreement of 0.25% is well within the stated accuracies of the two laboratories and is most gratifying [12].

In 1964, in response to the demand, tritiated toluene standards were prepared for the calibration of liquid-scintillation counting systems; these standards were relative to the water standards. The story [13] of their preparation affords a most instructive "case history" of the pitfalls that may be encountered in liquid-scintillation counting. In the course of developing these standards it was found that: commercial vials leaked to such an extent as to introduce quenching errors; white-capped vials gave up to 4.5% higher count rates than black-capped vials: from vial to vial there could be a systematic change as great as 3.5%: and mere rotation of a vial could introduce as much as a 1% change.

The effects of leaking vials are shown in Figures 14 and 15, the captions to which are self-explanatory. Figure 16 shows the count count-rate data for a group of eight different vials into which had been placed equal amounts of scintillator solution containing tritium. The liquid scintillator consisted of 5 g of 2,5-diphenyloxaxole (PPO) and 0.5 g of 1,4-bis-2-(4-methyl-5-phenyloxazolyl)-benzene (dimethyl POPOP) made up to 1 liter with toluene. This solution was diluted with ethyl alcohol in the proportion by volume of 70:30 and divided into 2 halves. To the one half was added quantitatively the NBS tritiated water standards together with inactive toluene: and to the other half the tritiated toluene sample together with triple-distilled water. In the identity of the two systems, this is similar to the internal standard identical system method of Moghissi, Hogrebe, and Carter [14,15], but without the internal standard.

Figure 14

Typical count-rate data on two vials selected from 35 vials and controls. No. 23 shows the effect of oxygen leaking in, while No. 35 was leak tight. At times A and B, the vials were momentarily opened. Each point represents the count rate for 100-sec counts, and the time between each reading for a given sample was about 70 min.

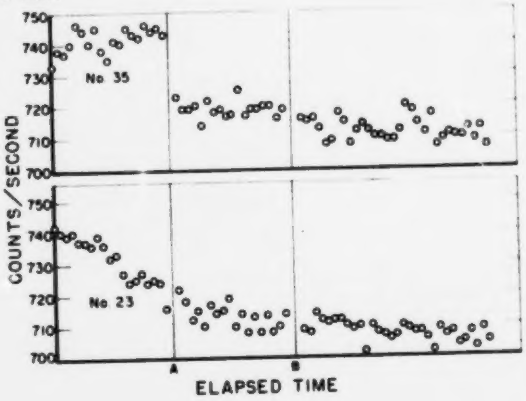


Figure 15

Typical count-rate data on three vials and on a flame-sealed ampoule (No. 13) used as control, selected from 15 vials and controls. The three vials had previously been flushed with nitrogen. No. 3 had a slow leak due to a faulty seal. No. 12 did not leak. No. 14 had a small hole drilled in the cap to provide a rapid leak. Each point represents the count rate for 100-sec counts, and the time between each point was about 35 min.

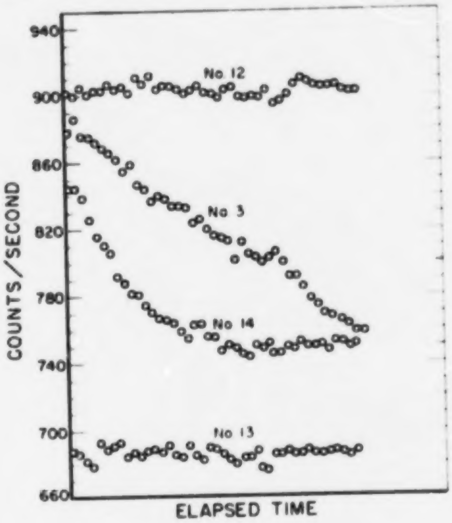
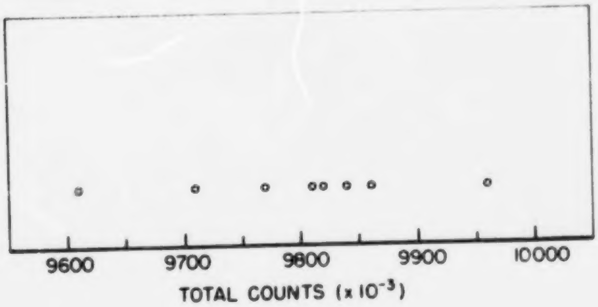


Figure 16

Typical data on a group of eight vials. Each point represents the total counts accumulated in ten 5-min readings. The total number of counts per sample was 10 million.



A group of 40 vials was then taken and their tops were ground flat. Teflon discs were inserted in the caps and a seal effected with a small amount of silicone grease. The scintillator solutions were allowed to stand in contact with silicone grease for 24 hours at 2°C to assure equal, if any, quenching due to the grease. The 40 vials were divided into 4 groups of 10 each. The first and fourth groups were each filled with about 15 ml of tritiated water in scintillator solution, and the second and third groups with tritiated toluene in scintillator solution. They were then all counted in an automatic sample changer for a large number of short durations each, in order to randomize rotational errors. An average count rate was then obtained for each group of vials which can be represented in arbitrary units as

$$W_1,\ T_1B,\ T_2B',\ W_2B'' \qquad (1)$$

where W_1 is the average activity in the first group of vials containing tritiated water, T_1 for the second group with tritiated toluene, and so on. B, B' and B'' are the average "bottle corrections" for the second, third, and fourth groups of vials relative to the first.

After this experiment the vials were all identified and washed with toluene, ethanol and ether and were found to have no memory effects. They were then refilled with the first group now containing tritiated toluene and so on, and recounted overnight, giving averages for each group that can now be represented by

$$T_1G,\ W_1BG,\ W_2B'G,\ T_2B''G \qquad (2)$$

where G is a possible "gain factor" between the two runs.

From the quantities shown in (1) and (2), we can obtain W_1/T_1B and T_2B'/W_2B'' from (1) and T_1/W_1B and W_2B'/T_2B'' from (2).

From these ratios $(W_1/T_1B)\ (W_1B/T_1) = (W_1/T_1)^2$

$$\text{and } (W_2B'/T_2B'')\ (W_2B''/T_2B') = (W_1/T_1)^2$$

On carrying out a number of such pairs of readings, values were obtained for W/T which were internally consistent with a standard deviation of about 0.1% but which differed between batches over a range of about 3.5%.

The method of weighing was suspected as being the cause of this final error and also possible evaporative losses in transferring the tritiated-toluene sample to the flask containing the liquid-scintillator solution. The weighing and evaporative losses were found to be very serious and accordingly a method was devised to obviate these, which is shown in Figure 17. The tritiated toluene and water samples were weighed into the small sample bottles and their weights followed for 10 minutes to assure that there was no evaporation. The two large flasks were weighed empty and then filled with appropriate amounts of liquid scintillator to give dilutions of the order of 1:1000. The small sample bottles were inserted into the larger flasks which were sealed, the small sample bottles being held at the top by means of a magnet acting on a ball bearing sealed into each of the small sample flasks. Upon release, the small sample flasks fell against the constriction in the neck of the flask, shown in Figure 17, opened, and the contents were then mixed. A further weighing gave the mass of scintillator plus sample, and hence the dilution factor. Further details are given in the referenced paper; in summary, however, this technique reduced the standard deviation in the comparative measurements to less than 0.1%. The final results for W/T are shown in Tables 1 and 2, and the certificate for the standards is shown in Figure 18.

Lastly, reference should be made to the measurements assurance program available from the National Bureau of Standards. We recently distributed some 25 samples of iron-59, under the sponsorship of the Nuclear Medicine Subcommittee of the Standards Committee of the College of American Pathologists, to medical laboratories to test their measurements procedures, the identity and activity of these samples being known only to the National Bureau of Standards. The results, which are in process of publication [15], are shown in Figure 19. We have also been requested to distribute calibrated tritiated water samples to four laboratories for a similar intercomparison of their measurements procedures.

Figure 17

Two flasks and sample bottles developed for an accurate dilution technique. The flask on the left has one of the sample bottles resting on the constriction. The flask on the right shows the botton half of the sample bottle "dropping" into the diluent.

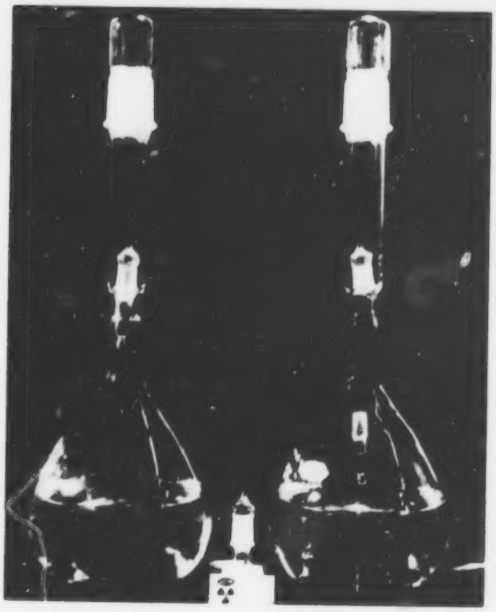
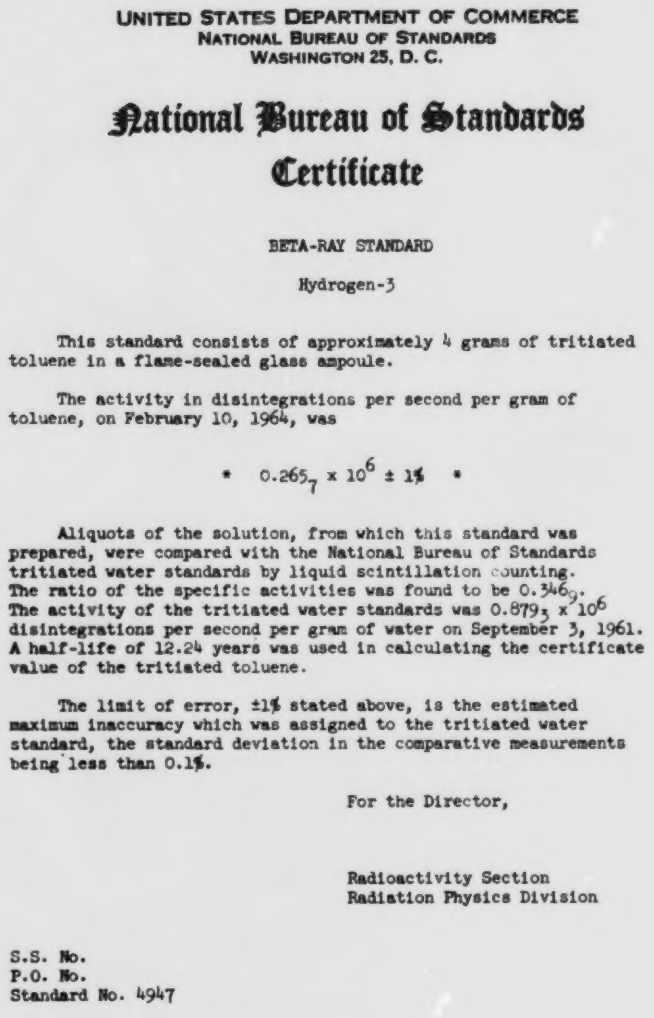


Figure 18

Certificate for the tritiated-toluene standard



UNITED STATES DEPARTMENT OF COMMERCE
NATIONAL BUREAU OF STANDARDS
WASHINGTON 25, D. C.

National Bureau of Standards
Certificate

BETA-RAY STANDARD

Hydrogen-3

This standard consists of approximately 4 grams of tritiated toluene in a flame-sealed glass ampoule.

The activity in disintegrations per second per gram of toluene, on February 10, 1964, was

* $0.265_7 \times 10^6 \pm 1\%$ *

Aliquots of the solution, from which this standard was prepared, were compared with the National Bureau of Standards tritiated water standards by liquid scintillation counting. The ratio of the specific activities was found to be 0.346_9. The activity of the tritiated water standards was $0.879_3 \times 10^6$ disintegrations per second per gram of water on September 3, 1961. A half-life of 12.24 years was used in calculating the certificate value of the tritiated toluene.

The limit of error, ±1% stated above, is the estimated maximum inaccuracy which was assigned to the tritiated water standard, the standard deviation in the comparative measurements being less than 0.1%.

For the Director,

Radioactivity Section
Radiation Physics Division

S.S. No.
P.O. No.
Standard No. 4947

Table 2

W/T ratios for four sets of vials for three sets of measurements. In any column, the ratios given in the first and third lines were obtained during one night's measurements, and the ratios in the second and fourth lines were obtained on the following night, the solutions and vials having been interchanged. The individual results for each night's reading for three sets of measurements are shown. In every case, W_1/T_1B is greater than W_1B/T and W_2B''/T_2B' is greater than W_2B'/T_2B'', indicating a finite vial error of between 0.3 and 0.6 percent.

	I	II	III
$\frac{W_1}{T_1B}$	2.8902	2.8921	2.8916
$\frac{W_1B}{T_1}$	2.8738	2.8685	2.8791
$\frac{W_2B''}{T_2B'}$	2.9082	2.9016	2.8799
$\frac{W_2B'}{T_2B''}$	2.8571	2.8834	2.8648

Table 3

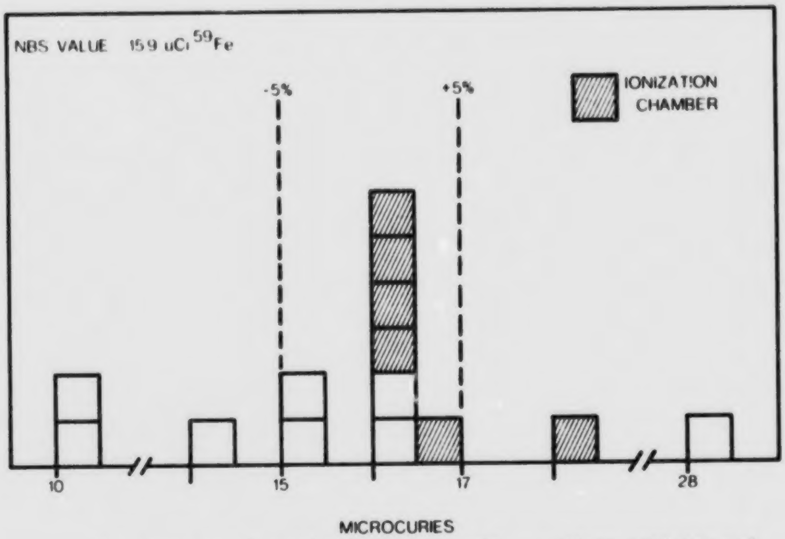
Final ratios of specific activities of tritiated-water standard to tritiated-toluene standard. The values of $\sqrt{W_1/T_1)^2}$, and $\sqrt{W_2/T_2)^2}$ are in close agreement, the standard error being 0.09 percent.

$\sqrt{\left[\frac{W_1}{T_1B}\right]\left[\frac{W_1B}{T_1}\right]}$	2.882	2.880	2.885
$\sqrt{\left[\frac{W_2B''}{T_2B'}\right]\left[\frac{W_2B'}{T_2B''}\right]}$	2.883	2.893	2.872

Figure 19

Results of ^{59}Fe activity measurement by 14 laboratories

CAP RADIONUCLIDE SURVEY



The function of standards is to provide a uniform national and international measurements system, so that measurements in one laboratory may be directly related to measurements in another laboratory anywhere else in the world. The advancement of science everywhere is based on the consistency of such a system, and we at the National Bureau of Standards do not consider that our mission is fulfilled merely by the issuing of standards. Only when concordant measurements based on these standards come back from other laboratories do we feel that we have succeeded in our mission. There is, therefore, a very strong motivation on our part to assist to the utmost of our ability in any "round robin" comparisons that support and strengthen the national and international measurements systems.

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DETERMINATION OF TRITIUM IN AN ANALYTICAL CHEMISTRY LABORATORY

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The quantitative analysis of tritium in an analytical chemistry laboratory exhibits various aspects, depending on experimental objectives and the type of analysis attempted. In addition to conventional methods of gas analysis, various other techniques are available, based on the radioactivity of tritium. The analyst is, thus, required to choose the most suitable of several techniques, using criteria such as the degree of accuracy required, response time, cost of analysis, sample quantities available, etc.

This paper relates the experience of a chemical laboratory working in this area over the past few years. Four classes of products analyzed in the laboratory, and the measurements applied to each, are given in Table 1.

Table 1

Measurements Applied to Tritiated Compounds

Pure tritium	isotope ratios impurities % of tritium
Tritium-based gas mixtures	% of tritium isotope ratios
Gas or liquid mixtures containing traces of tritium	% of tritium
Various substances containing tritium	% of tritium

The following analytical methods may be employed: Gas-phase chromatography, mass spectrometry, measurement in an ionization chamber, calorimetry and liquid scintillation counting. These methods are briefly discussed and compared, and the analytical method used by the laboratory for routine checks is specified.

Gas Chromatography

The separation of the six isotopic compounds of hydrogen was studied by many workers [1-9]. However, the purpose of the previous studies was mainly qualitative or preparative in character. In order to get a quantitative analysis which can be used for routine checks, the method has to fulfill the following requirements:

The peaks must be tapered and symmetrical to allow accurate measurement of height and area.

The resolution factors must be high enough to avoid overlapping.

The retention times must be short.

The sensitivities must be high for each compound.

For these reasons a short column with a deactivated stationary phase was chosen. A column three meters long filled with alumina covered by ferric hydroxide and partially deactivated by carbon dioxide gave the best results.

Neon was used as carrier gas because its thermal conductivity is very different from that of elemental hydrogen and allows good sensitivity for all the isotopic compounds and ^{3}He. The gas chromatography was performed at liquid nitrogen temperature and analysis was carried out with a double-detection system consisting of a catharometer and an ionization chamber.

The catharometer allows the determination of helium-3 and the six isotopic compounds $^1H_2$ - ^1HD - ^1HT - D_2 - DT - T_2. However, stabilization is time-consuming, response factors are different for the six peaks, and calibration is difficult to make because substances such as ^1HD, ^1HT, and DT need to be specially prepared for this purpose. A chromatogram obtained by this procedure is shown in Figure 1.

Detection by the ionization chamber only allows determination of the tritiated molecules ^1HT, DT, and T_2. However, stabilization is rapid and detection limits are very low. A typical chromatogram is shown in Figure 2.

Figure 1

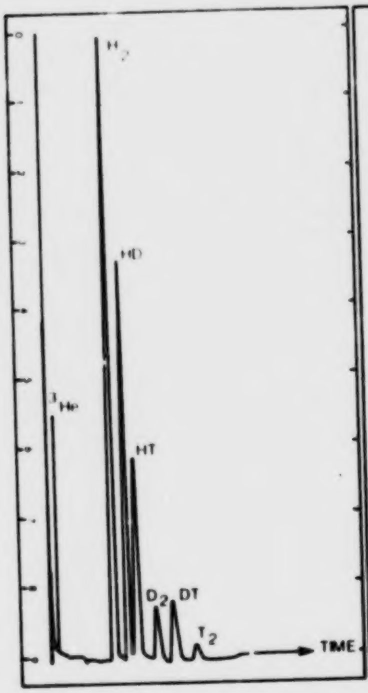
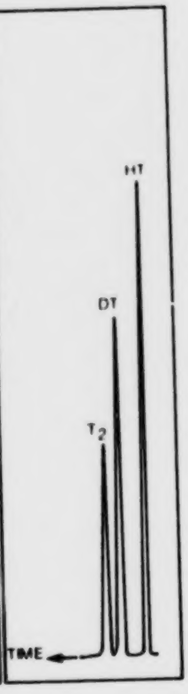


Figure 2



Once the concentrations of the three tritiated molecules are known, the desired isotopic ratios may be determined, provided that the mixture was equilibrated before analysis. The six isotopic compounds of hydrogen are governed by three equilibrium relations to each of which corresponds an equilibrium constant depending exclusively on temperature.

$$^1H_2 + D_2 \rightleftharpoons 2\,^1HD \qquad K_1 = [^1HD]^2/[^1H_2]\,[D_2]$$

$$D_2 + T_2 \rightleftharpoons 2\,DT \qquad K_2 = [DT]^2/[D_2]\,[T_2]$$

$$T_2 + {}^1H_2 \rightleftharpoons 2\,^1HT \qquad K_3 = [^1HT]^2/[^1H_2]\,[T_2]$$

Bombardment with tritium beta particles tends to equilibrate these mixtures, but the reaction is not immediate. On the other hand, experiments show that by heating the gas mixture with a red hot tungsten filament a state of equilibrium is obtained which corresponds to a temperature depending on the experimental installation employed.

Since the response of the ionization chamber depends exclusively on the quantity of tritium and not on the nature of the substance, by calibrating the chamber with one of the tritiated molecules the concentrations of ^1HT, DT and T_2 may be determined by integration. The concentrations of $^1H_2$, ^1HD, and D_2 may then be easily deduced by using the constants K_1, K_2, and K_3.

This procedure nevertheless requires calibrations of the ionization chamber and measurement of the volume of gas injected. Experience has shown that accuracy and especially speed can be improved if calibration and volume measurement can be avoided. The desired isotopic ratios may be determined once the ratios of the areas of the peaks ^1HT and T_2, and DT and T_2 are known.

Let A and B be respectively the area ratios S_{HT}/S_{T2} and S_{DT}/S_{T2}. The isotopic ratios required will be:

$$^1H/(^1H + D + T) = \frac{A\left[2B\sqrt{\frac{K_1}{K_2K_3}} + \frac{4}{K_3}A + 1\right]}{4\left[\frac{A^2}{K_3} + \frac{B^2}{K_2} + AB\sqrt{\frac{K_1}{K_2K_3}}\right] + 2A + 2B + 1}$$

$$D/(^1H + D + T) = \frac{B\left[2A\sqrt{\frac{K_1}{K_2K_3}} + \frac{4}{K_2}B + 1\right]}{4\left[\frac{A^2}{K_3} + \frac{B^2}{K_2} + AB\sqrt{\frac{K_1}{K_2K_3}}\right] + 2A + 2B + 1}$$

$$T/(^1H + D + T) = \frac{A + B + 1}{4\left[\frac{A^2}{K_3} + \frac{B^2}{K_2} + AB\sqrt{\frac{K_1}{K_2K_3}}\right] + 2A + 2B + 1}$$

In this case, the catharometer, with its long stabilization time, low sensitivity and complicated calibration requirements, may be abandoned, except for determinations of helium-3. The use of neon as a carrier gas permits the determination of small quantities of helium-3 (detection limit 0.1%).

By the use of a digital integrator, the surface area values of the three tritiated peaks may be transmitted to punched tape. By feeding this information, together with the equilibrium constants, into a WANG 700 computer connected to a typewriter, the analytical results are automatically printed out.

After a series of nine measurements carried out in the same mixture of T, D, and ^1H the percent isotopic ratio was found to be 21.23±0.26, 77.30±0.29, and 1.46±0.03, respectively at 99% confidence level.

The overall accuracy of results obtained is hard to evaluate, since it is affected by the uncertainty of the constants and the conditioning of the samples. A fair estimate is two to three percent. In case of high purity tritium, the accuracy estimate is ±0.1% as shown in Table 2.

Table 2

Isotopic Ratios of High Purity Tritium in Percent

T	D	^1H
99.53	0.17	0.30
99.75	0.20	0.05

A remark is appropriate here concerning the problem of tritium storage. Besides the well-known phenomena of adsorption on the walls, the presence of grease, especially on ground sockets, causes a very drastic decline in the tritium content in the long run.

The quantitative analysis of impurities in tritium is necessary for the determination of the tritium percentage of the gas used for calibration, when a high degree of accuracy is required. The analysis is then carried out in two steps:

1. The gaseous impurities (Σ imp) are first determined. Thus, $(^1H + D + T)\% = 100 - \Sigma \text{ imp}$

2. Isotopic analysis leads to the determination of the ratio $^1H{:}D{:}T$. The percentage of tritium can then be deduced as: $T\% = T(100 - \Sigma \text{ imp})/(^1H + D + T)$.

The following gaseous impurities are found in tritium: nitrogen, oxygen, methane, argon, and carbon dioxide.

Separation was carried out in two switchable columns mounted in parallel. The first was a molecular sieve column, separating out oxygen plus argon, nitrogen, and methane. The second column was filled with porapak Q and permitted determination of the carbon dioxide content. The separation of oxygen from argon was carried out at -80°C.

Detection was effected by catharometer; an ionization chamber placed in series indicated the quantity of tritium present in the methane in the form of tritiated methane. It has been shown that there is no oxygen in gas containing a high percentage of tritium; oxygen reacts quickly with tritium to give tritiated water.

Mass Spectrometry

Mass spectrometry is one of the most powerful methods for hydrogen isotope analysis, and many studies have been made on this subject [10-15]. Most of these studies concern mixtures containing one or more compounds at trace level.

When none of the compounds are at trace level, the analysis of the six isotopes of hydrogen poses a difficult problem due to the large number of ions present and the variety of their sources, as shown in Table 3.

Table 3

Repartition of Ions in the Different Peaks

MOLECULAR WEIGHT / MOLECULES	1	2	3	4	5	6
$^1H_2$	$^1H^+$	$^1H_2^+$				
^1HD	$^1H^+$	D^+	$^1HD^+$			
^3He			$^3He^+$			
^1HT	$^1H^+$		T^+	$^1HT^+$		
D_2		D^+		D_2^+		
DT		D^+	T^+		DT^+	
T_2			T^+			T_2^+

It may be seen that monoatomic ions are each derived from:

$^1H^+$ from $^1H_2$, ^1HD, or ^1HT

D^+ from D_2, ^1HD, or DT

T^+ from T_2, ^1HT, or DT.

Also, if difficult to use atomic weight 1 is eliminated, only molecular weights 5 (DT^+) and 6 (T_2) are free from interference. One gets five relations (Masses 2 - 3 - 4 - 5 - 6) with seven compounds (hydrogen isotopes plus ^3He).

Two methods may be employed:

1. Equilibration of the mixture and use of the equilibrium constants K_1, K_2, and K_3 defined above, which give three more relations and allow solving the system. But the gas mixture must be equilibrated before analysis;
2. The use of the "fine" slits of the instrument, permitting resolution of the peaks ^1HT and D_2 for molecular weight 4, and peaks He + T and ^1HD for molecular weight 3. (The peak He + T can only be split by a spectrometer with a resolution power of 150,000 or greater).

The use of "wide" slits (Figure 3) (low resolution, high sensitivity) produces peaks for molecular weights 2, 3, 4, 5, and 6, and the fine slits (Figure 4) (high resolution, low sensitivity) provide the ratios $^1HT/D_2$ and $(He + T)/^1HD$. This last method, which allows obtaining the individual concentrations, was chosen as a check method.

Figure 3

Mass Spectrometry, Wide Slits.

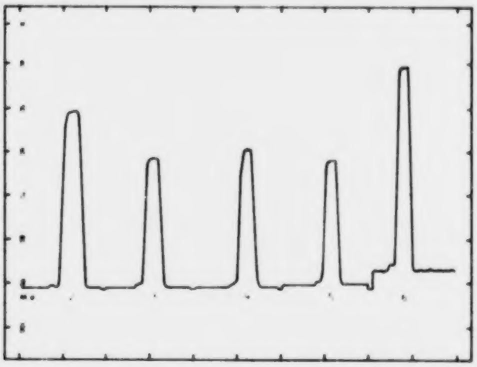
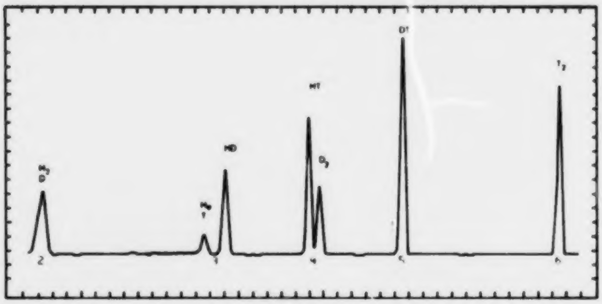


Figure 4

Mass Spectrometry, Fine Slits



To determine the concentrations, it is necessary to calibrate the mass spectrometer for each compound and to calculate the contributions on masses 2 (D^+) and 3 (T^+). It has been shown that with a fine adjustment of the Varian Mat CH_4 mass spectrometer, the sensitivities of $^1H_2$, D_2 and T_2 are nearly the same. It has been verified that the sensitivity for ^1HD is the arithmetic mean between $^1H_2$ and D_2. So the sensitivities for ^1HT and DT were considered as equal to the arithmetic means between $^1H_2$ and T_2 for ^1HT and between D_2 and T_2 for DT. The calibration curve for He was established separately with ^3He. The ratio of sensitivity $^3He/^4He$ was measured and ^4He is now currently employed for calibration.

The contributions of $^1H_2$ as ^1H, D_2 as D and T_2 as T are small ($D/D_2 = 4.10^{-3}$) and not different. The contribution of ^1HD as ^1H and D is about half the contributions of $^1H_2$ as ^1H and D_2 as D. The same rule was used for the contribution of ^1HT as ^1H and T and for DT as D and T.

Mass 6 peak gives T_2 and permits calculation of the contribution of T_2 as T on mass 3 peak. Mass 5 peak gives DT and permits calculation of the contribution of DT as D on mass 2 peak and as T on mass 3 peak. Mass 4 peak wide slits give the sum $^1HT + D_2$. Mass 4 peak fine slits provide the ratio $^1HT/D_2$. These measurements permit calculation of ^1HT and D_2 and the contribution of ^1HT as T on mass 3 peak and of D_2 as D on mass 2 peak. Mass 3 peak wide slits give the sum $^1HD + {}^3He + T$ and mass 3 peak fine slits provide the ratio $^3He + T/^1HD$. As T (contribution of ^1HT, DT, and T_2 on mass 3 peak) was calculated previously, it is possible to get ^1HD and ^3He, and to calculate the contribution of ^1HD as D on mass 2 peak. Mass 2 peak gives the sum $^1H_2 + D$. The contribution of ^1HD, D_2 and DT on mass 2 peak, D, has been calculated previously, so it is possible to get $^1H_2$.

By employing a certain number of experimentally justifiable approximations, the problem is considerably simplified. This method gives results that are accurate to ±3%.

The results of a comparison between gas chromatography and mass spectrometry are given in Table 4.

Table 4

Comparison Between Mass Spectrometry and Gas Chromatography

Isotope Ratio in %	Mass Spectrometry	Gas Chromatography
T	20.6	20.3
D	77.9	78.2
^1H	1.5	1.5

Measurement in an Ionization Chamber

Measurement in an ionization chamber is used to check the calibration mixtures and to make sure that the value measured by counting is not very different from the theoretical value. This conventional technique has been widely described. Two procedures are available: direct measurement, and measurement using a calibrated instrument.

Direct measurement consists of estimating the quantity of tritium on the basis of the physical constants of the gas and the intensity of the radiation passing through the chamber. Due to the uncertainty surrounding the physical constants, the exact volume of the chamber and the calibrated resistance, it is not possible to obtain results closer than ±5%. Though attractive in many ways, this procedure was therefore abandoned in favor of measurement using a calibrated instrument [16].

After a series of 12 measurements carried out on the same gas mixture, a tritium value of 19.95 ± 0.08% at 99% confidence level was obtained.

The relative accuracy of routine checks depends on the accuracy of calibration of the chamber (tritium content, pressure measurements). The estimate is ±2%.

Calorimetry

Due to the low penetrating power of tritium beta particles, all the energy dissipated by a tritiated substance enclosed in a given space is transformed into heat. By measuring the heat flow and employing certain known physical constants of tritium, the tritium content of the sample may be determined [12,17,18]. The main difficulty confronting this method lies in the calibration. Several techniques are available, including the Joule effect and the use of a well-known radioactive compound emitting alpha or beta particles. Calibration by the Joule effect is marred by heat leakage along the heater wire. When a radioactive substance, e.g., plutonium, is used for calibration, errors may be caused by uncertainty concerning the abundance of isotopes 238, 240, and 241 and the presence of americium-241.

Due to the systematic deviation between these two calibrations, the mean of the two values is employed. The ideal solution would be to calibrate the instrument with tritium; however, due to the limited dimensions of the measurement cell, the volume of gas introduced is insufficient to produce a signal of adequate amplitude. The tritium would have to be introduced in condensed form. Tests are currently being conducted with tritiated uranium.

After repeated measurements on the same substance, a tritium value of 13.56 ± 0.04 mg at 99% confidence limit was obtained.

Liquid Scintillation [19-22]

The analysis of gases for traces of tritium particularly in helium-3, was carried out with a liquid scintillation counter. Tritium is oxidized on a metal filament covered with copper oxide and heated to 450°C by the Joule effect.

The oxidation of about 15 ml of gas takes place in a sampling flask which is also used as measuring flask. The transfers which generally lead to important losses are avoided by this process.

In the absence of a standard, only the reproducibility of the method can be estimated. After a series of eight analyses, a tritium value of $(3.2 \pm 0.7)10^{-8}\%$ was obtained using the liquid scintillation technique. The same mixture yielded a value of $<8 \times 10^{-8}$ using an ionization chamber.

Two tests carried out with helium theoretically containing $1.6\ 10^{-10}\%$ T yielded values of $3.6\ 10^{-10}$ and $3.1\ 10^{-10}\%$, which can be considered a satisfactory approximation at this concentration.

Conclusion

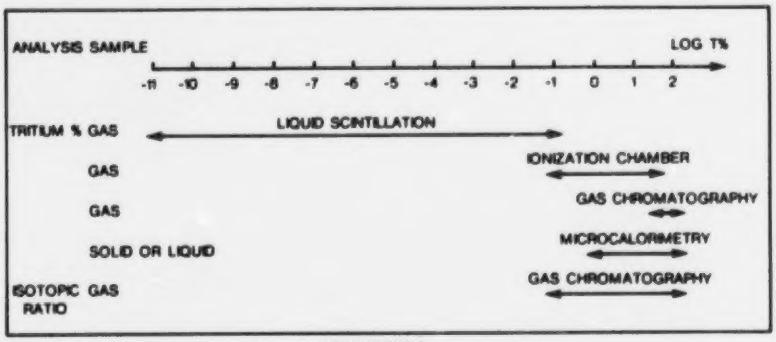
The question finally requires the choice of a method. No single method is universally valid, and the choice for routine measurement must be made on the basis of conditions prevailing in the particular laboratory.

For the determination of isotope concentrations and inlet gas impurities, gas chromatography was chosen as the most suitable procedure in preference to mass spectrometry. Gas chromatography requires relatively simple equipment which may be assembled in the laboratory, whereas the mass spectrometer is very delicate and less dependable. The gas chromatograph is about 20 times cheaper than a mass spectrometer, resulting in vastly reduced capital outlay.

The gas chromatograph is also far more flexible than the mass spectrometer, and may be used without calibration as demonstrated above. Mass spectrometry was abandoned for routine checks but is used for qualitative analysis problems.

For the other measurements, Figure 5 shows the method of analysis chosen according to the nature of the compound analyzed and its tritium concentration.

Figure 5



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GAS COUNTING OF TRITIUM

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The difficulty in measuring tritium results from the low-energy of the beta particle, a maximum energy of 18 keV emitted by the spontaneous decay of tritium. Because of this low energy, the most appropriate method of measurement is by intimate mixture of the sample with the detection medium. Thus, both internal gas counting and liquid scintillation counting are used. The purpose of this paper is to review information on techniques of tritium measurement by gas counting and to indicate how some practical problems of calibration and standardization can be simplified by the use of appropriate procedures. The author's preference for techniques to measure low levels of radioactivity will be given greater emphasis, but an evaluation of other techniques also can be obtained from the text and from the literature references.

The type of gas counting, whether by ionization, proportional or Geiger counter, can be defined by the anode potential; a) Ionization region: at a low voltage, in a suitable gas, electrons and positive ions migrate to the electrodes and transfer an elementary charge. The collected charge measures the number of ionization events and energy induced in the counting gas by the primary radioactive decay; b) Proportional region: as the voltage is increased beyond the region of simple ionization into the proportional counting region, the electrons moving toward the anode are exposed to a strongly increasing electric field, causing secondary gas ionization; the secondary electrons produced repeat the process, as the voltage is increased, and gas multiplication occurs. The primary ionization can be amplified 10^3 to 10^5 times, depending on the counting gas, and the pulse output is proportional to the energy of the initial radioactive decay; c) Geiger region: as the voltage is increased further into the Geiger counting region, all ionizations are multiplied to the same pulse size and energy discrimination is lost. The discharge occurs along the whole of the anode wire and strongly resembles an instantaneous glow discharge [1,2].

Measurement by Ionization Chamber

An ionization chamber for tritium measurements can be simple. A sensitive electrometer is required to measure the small current produced. The calibration of a given ionization chamber is usually linear, and the limitations of instrumental background depend on the type of electrometer used and the leakage current of the insulators. The theory and design of electrometers and the properties of insulators suitable for ionization chambers are reviewed by Fairstein [3]. Additional details and references on ionization chambers and pulse ionization chambers for energy discrimination are reported by O'Kelley [4], Jacobs [5] and Soudain [6].

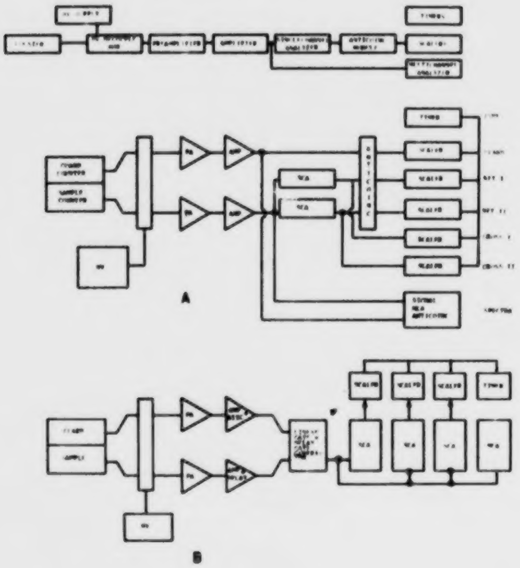
In the tritium ionization chamber described by Tolbert [7], a background of 2.5 X 10^{-17} amps would give a limiting detection of approximately 500 dpm/g H_2O. The same ionization chamber would be suitable for tritium measurements that are many orders of magnitude higher than this, depending on the electrometer used.

Proportional Region Counting

Tritium in the proportional region can be counted with a low background and high sensitivity. Pulse output can be amplified to achieve energy discrimination; gas multiplication produces pulses larger than those of the ionization chamber and thus can be more easily amplified by a preamplifier and amplifier for counting. A block diagram of such a system is shown in Figure 1. Proportional region counting can measure events at high count rates without significant losses because of the small detector dead time.

Figure 1

Electronic components and logic for proportional counting systems.
- a. Standard anticoincidence logic used in most existing systems.
- b. Logic employing anticoincidence with timing signal delay and delay gate generator.



Geiger Region Counting

Tritium counting in the Geiger region requires simpler electronic equipment than does proportional counting. Primary differences are in the counting gas and applied voltage; each ionization results in a Townsend avalanche and a large discharge at the collector wire. Simple amplifiers and scalers are suitable for recording the events. One problem with the Geiger-region discharge is that there is a time interval during which the tube is insensitive to ionizing particles (dead time). For high counting rates this time, 100 to 300 μsec, becomes significant, and corrections for coincidence loss must be made. Since every ionization produces a pulse of the same size, energy discrimination is not possible. Thus, reduction in background must be made exclusively by shielding, both massive and with an anticoincidence detector. Detailed treatment of this subject is given by Wilkinson [2] and Koroff [8]. A practical survey of Geiger-Müller counting is found in O'Kelley [4].

Proportional or Geiger counters for tritium are simple in construction, consisting of a metal or metallized quartz cylinder cathode (50 mm diameter) and an insulated center-wire anode (ca. 0.025 mm). A high voltage is usually applied to the center wire. Such tube designs are given by Ostlund and Werner [9], O'Brien [10] and Geyh [11]. Hydrogen cannot be counted alone in the Geiger region. However, with a few torr propane and an external electronic quenching circuit, the counting properties were found to be excellent [9]. In the Ostlund system, 750 torr hydrogen and 10 torr propane are used. O'Brien [10] reviews the counting of tritium in the Geiger region and especially the properties of some normally used quenching gases. Small amounts of ethylamine or toluene were found to be suitable as quenching gases for counting tritium prepared as hydrogen at pressures of 500 torr. One major problem in Geiger counting is, as mentioned previously, that it is impossible to discriminate between the decay of alpha-emitting radon and that of tritium. Environmental radon is a common contaminant when tritium samples are prepared and it should be monitored. If the time between sample preparation and counting can be about three weeks, the contamination from radon (half life of 3.8 days) would be essentially eliminated.

Preparation of Samples

Requirements for the preparation of the gas differ for each type of counting. The sensitivity required and radioactive concentration needed to measure a signal above background radiation are most important variables; for the measurement of low tritium concentrations, the natural background radiation must be limited.

Since ionization chambers measure the current flow induced in an electrode by the decaying radionuclide, the requirements for gas composition are minimal. Thus, sample preparation is also minimal. Survey meters for tritium are often ionization chambers that pass a known flow rate of air through a given chamber volume, the main sample preparation being the removal of dust particles. Background can be reduced by external gamma radiation compensation [12]. The level of sensitivity is appropriate for certain reactor uses, tracer applications, and monitoring instruments in fuel reprocessing laboratories. The detection range of a large Kanne chamber [13,14] is in the region 10^{-5} to 60 μCi/ml. Commercial instruments [49] are available with sensitivity of ca. 130 pCi/g H_2O (29,000 dpm/g). For increased sensitivity it is important to devote more effort to sample preparation.

The most critical requirements for purity of gas are in proportional counters, and thus significant effort must be given to the preparation of a sample gas that is "pure," i.e., free from electronegative impurities. In proportional and Geiger counters, it is common to reduce the water to hydrogen. In several techniques hydrogen is counted directly and in others it is converted into hydrocarbons by appropriate catalysts.

In all sample preparation techniques, the rule is to minimize handling of the sample. Thus, the simplest technique is often the most reliable, with limited problems of sample contamination and losses. Table 1 is a summary of the techniques of sample preparation for gas counting.

Table 1

Preparation of Counting Gas for Tritium

Hydrogen Preparation

$$H_2O + Mg \xrightarrow{600^oC} MgO + H_2$$

$$H_2O + Zn \xrightarrow{400^oC} ZnO + H_2$$

$$H_2O + U \xrightarrow{800^oC} U_nO_m + H_2$$

Hydrocarbon Preparation

$$SrC_2 + 2H_2O \longrightarrow C_2H_2 + Sr(OH)_2$$

$$C_2H_2 + 2H_2 \xrightarrow[50^oC]{Pd} C_2H_6$$

$$C_2H_4 + H_2 \xrightarrow[50^oC]{Pd} C_2H_6$$

$$CO_2 + 4H_2 \xrightarrow[375^oC]{Ru} CH_4 + 2H_2O$$

$$CO_2 + 2H_2O + 4Zn \longrightarrow CH_4 + 4ZnO$$

$$Al_4C_3 + 12H_2O \longrightarrow 4Al(OH)_3 + 3CH_4$$

$$C_3H_4 + 2H_2 \xrightarrow[50^oC]{Pd} C_3H_8$$

$$CH_4 + HT \xrightarrow[350^oC]{Ru} CH_3T + H_2$$

Because of the properties of the hydrogen and of the tritium molecule, i.e., small size and reactivity, elemental tritium is readily lost, absorbed and exchanged with materials with which it comes in contact. The materials used to construct gas handling lines and detectors must be chosen to minimize these effects. If samples of both high- and low-level radioactivity are to be analyzed, separate systems should be used. After a high-level sample is prepared, background samples are consistently contaminated. To eliminate the contamination, it is necessary to clean all grease from the lines, as stopcock grease has been found to retain elemental tritium. In all-metal lines without grease, contamination may still occur by the diffusion of tritium into the interstices of the metal and from outgassing of rubber "o" rings. The contamination of subsequent samples can often be reduced sufficiently by flushing the lines several times with "tank" hydrogen and by heating them. It is the author's experience that metal lines are less susceptible to contamination and are more readily decontaminated. Since decontamination is not easy to accomplish in either system, careful selection of the samples to be measured in a given system is most desirable.

Figure 2

Sample preparation and counter fill manifold.

- a. Heating mantle
- b. Magnesium furnace
- c. Glass "U" tube
- d. Stainless steel traps filled with activated charcoal

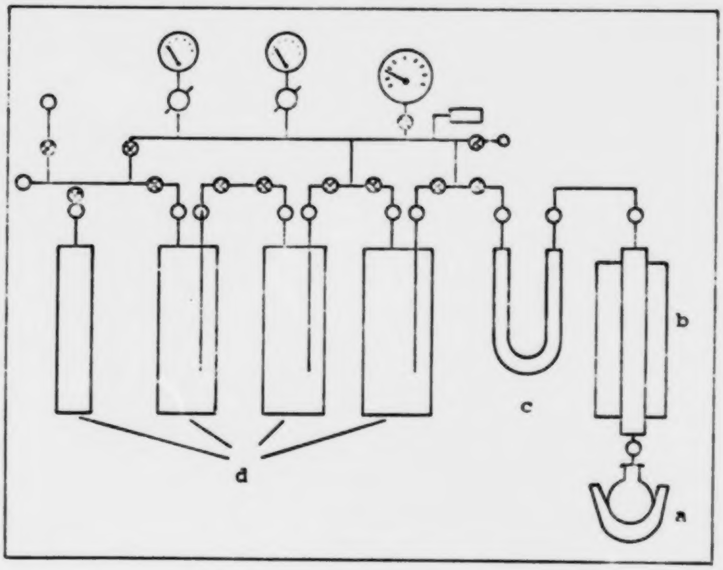


Figure 2 shows a simple metal manifold constructed at the International Atomic Energy Agency, Vienna, for the reduction of water samples with magnesium and the collection of the hydrogen on an adsorbent trap. A small trap that permits the addition of hydrogen to the detector together with a counting gas is included. The counter-filling system can operate up to pressures of 7,600 torr. It is used routinely at hydrogen counting pressures of 3,500 torr. The details of two similar gas-preparation systems can be found in Schell [15,16]. The procedure used for preparation of the samples is as follows:

> The system is degassed by heating the activated charcoal-filled traps, magnesium furnace and lines while evacuating. A water sample is inserted at the base of the furnace and the air above the water is removed rapidly by evacuation. Dry ice is used to cool the glass "U" tube for collection of water which may not have been reduced completely; liquid nitrogen is used to cool the stainless steel traps filled with charcoal for the collection of hydrogen. The water is evaporated slowly and the hydrogen gas which passes through the cooled glass "U" tube is collected in the adsorbent trap.

The time required to process each sample is about 30 minutes when the lines and traps are already prepared. With the capacity of this magnesium furnace, it was found that four samples could be processed with a single charge of magnesium. The glass "U" tube permits visual observation of the water that may not have been reduced in the furnace. With samples of approximately the same tritium concentration, negligible contamination was found. A consistent and satisfactory procedure is to measure the high-level sample or standard as the last sample and background radiation or a low-level sample as the first sample. With expected differences in sample concentration greater than 10^4 TR, a new charge of Mg is required.

A system for processing water samples and converting the hydrogen into ethane is one of the more popular methods of sample preparation for gas counting [17]. In this method, water is reduced by Mg, Zn or U, and the hydrogen is collected on a molecular sieve trap cooled to liquid air temperature. Acetylene or ethylene prepared from petroleum, so it is tritium free, is condensed with liquid air into a bulb (ca. 5 liters) containing a Pd catalyst (Englehard Industries, Neward, NJ). The sample hydrogen is expanded from the molecular sieve trap by being heated and is allowed to react with the ethylene in a 5-liter glass reaction bulb. Approximately 10 percent excess ethylene is added, which allows the reaction to proceed to completion (i.e., all sample hydrogen is utilized) within two hours. By using liquid air, the sample can then be transferred easily into storage vessels or into a detector without sample loss, because of the higher boiling point of liquid ethane of -88.3°C. The counting gas mixture consists of the excess ethylene and sample hydrogen as ethane. The acetylene or ethylene tanks obtained from a supplier should be checked for possible tritium contamination. Since all counting measurements are relative and the background radiation is subtracted, it is possible to correct for the contamination although the true counter background is not measured. Both ethylene and ethane are excellent counting gases and can tolerate some air impurities without their counting characteristics being changed.

One problem that has been encountered during several years of ethane counting is the contamination of background gas by repeated counting of the same background sample. It is necessary to make up and count new background samples frequently to avoid a systematic increase in background count rate. In a low-level laboratory, the increase in count rate is only slightly greater than the statistical deviation of the background counting error. However, the count rate has been found to increase systematically with time and number of times the same background sample is counted. This problem is important only when samples with a low tritium concentration are measured. The error can be minimized by counting the same background gas not more than three times. For routine work the ethane method is quite satisfactory.

Another contamination problem has been found with the catalyst. The Pd catalyst probably collects tritium during or after its manufacture, since the first sample to be measured on a new catalyst is often highly contaminated. In fact, the contamination has been found to be great enough to contaminate an entire glass preparation system. A routine procedure adopted at the IAEA has been to heat and evacuate each batch of catalysts with several flushings of tritium free hydrogen before use.

The contamination of the laboratory from workers' luminous watches, which release tritium, is another potentially serious problem [18].

The most desirable molecule for gas counting is the one with the largest number of hydrogen atoms. Methane, ethane and propane are suitable as tritium counting gases for this reason [19-21]. The use of methane as a counting gas is most desirable in low-level counting, since high pressures and thus large amounts of hydrogen can be measured at low background [22]. Tritium can be incorporated in the methane molecule by one-step synthesis. The reaction chamber consists of two heated zones, one for the Ru catalyst (Englehard Industries, Newark, NJ) at 380°C, and one for the Zn metal at a temperature just below the boiling point of Zn, 400 to 410°C [23]. The reaction chamber can be loaded and permitted to react unattended overnight.

One problem with this technique is possible environmental contamination, since the chamber must be opened to the atmosphere after each sample is prepared, because of Zn oxidation during the reaction. The chamber must be evacuated well before the next reaction. The author tried this procedure using a slightly modified design and found the reaction proceeded to completion in a few hours. However, it was not possible to avoid contamination. Tritium free background water consistently had ca. 40 TR or greater after the reaction. Additional efforts to improve this technique by reducing contamination would be worthwhile.

Another technique for the preparation of methane is by reacting aluminum carbide with water. From the reaction in Table 1, it can be seen that part of the hydrogen (tritium) is tied up in the product, aluminum hydroxide; thus some losses and fractionation occur. To obtain reliable results with the aluminum carbide reaction, this loss must be corrected.

The alkaline earth metal carbides are used to produce acetylene by hydrolysis. Of these carbides, SrC_2 is the one most commonly used. The acetylene so produced in the reaction can be measured directly or it can be converted into ethane by catalytic hydrogenation with the catalyst Pd on asbestos. The SrC_2 must be kept dry and free from atmospheric moisture to limit contamination. The $Sr(OH)_2$ produced in the reaction shown in Table 1 contains some tritium and, again, isotopic fractionation would be a problem. This problem is circumvented by Suess, who routinely produces acetylene from tritium free water. The gas is stored for later sample preparation of ethane by the Bainbridge methods [20,24].

There are numerous exchange reactions in which sample hydrogen can be used to label organic gases such as methane, ethane and propane for gas counting. A simple procedure to label methane is as follows:

> Sample water is reduced to hydrogen and added to a storage vessel. A quantity of methane is mixed with the hydrogen at an optimum ratio 4.5:1,H_2:CH_4 and circulated with a small membrane pump over a Ru catalyst (Englehard Industries, 5 percent Ru on Al_2O_3 pellets). After one hour at 350°C the calculated exchange has occurred and the hydrogen and tritiated methane can be separated on a charcoal trap cooled with liquid nitrogen for counting [25,26].

Organic samples can be prepared for gas counting by pyrolytic reduction in the presence of water by Zn and NiO [27]. A modification of this technique permits both ^{14}C and ^{3}H to be measured in the same sample by gas counting in the proportional region [28].

Enrichment Techniques for Gas Counting

One technique for sample preparation is low temperature gas chromatography. With this technique, significant enrichment of tritium from hydrogen can be achieved. Water samples are first reduced to hydrogen and then added to a cooled adsorbent column. Because of the differences in adsorption of the $^{1}H_2$, ^{1}HT, and T_2 molecules, separation by elution can be achieved [29,30,31]. In the Hayes and Hoy procedure, an automated Pd gas chromatograph is reported having a capacity of 50 liters of hydrogen, i.e., 40 ml water equivalent. For two samples per day in the range of 5 to 150 TR, the system yields reproducible results, with an efficiency of 81.2±1.6 percent for tritium recovery. Some problems in such a system are initial cost, construction difficulties, and the limited number of samples measured per unit time.

Thermal diffusion techniques of enrichment have been used successfully to concentrate tritium from hydrogen [32,33,34]. In this technique, hydrogen is permitted to diffuse through Pd metal into a series of columns. Each column consists of a central wire heated to ca. 1,200°C and a cold jacket. Tritium is enriched at the cold wall and carried downward by convective currents because of its greater density; it is removed from the column by a Toeppler pump. Enrichment of 100 to 500 times can be accomplished in one to two days. When the system is set up, little worker time is necessary to run the samples and virtually no contamination is possible because it is an enclosed system. The problems lie in the initial construction and limited number of samples per system in a unit time.

The most common method for preparing low-level samples is by electrolytic enrichment [9,35-39]. In this technique, a series of cells can be set up for simultaneous enrichment of many samples with known calibration standards. The tritium enrichment factor can be estimated from the enrichment of the natural deuterium of the sample or by measuring a tritium standard before and after enrichment. Since the current is identical in all samples run in series, the enrichment factor can be applied to the unknown samples. Problems with the determination of the enrichment factor are differences in individual cell properties and temperature gradients in the coolant bath. In the modified Bainbridge system at the IAEA, a single-stage enrichment factor of 18±0.5 is obtained routinely in 700 ampere hours with an elapsed time of four days. Two systems, one twenty-cell and one sixteen-cell, are operated at ca. 0°C with a recovery of ca. 80 percent of the tritium. Such a system permits large numbers of samples to be processed routinely. The enriched water is then converted into a suitable gas for counting.

Low Background Gas Counting Techniques

In the detection of low levels of tritium some important factors are as follows:

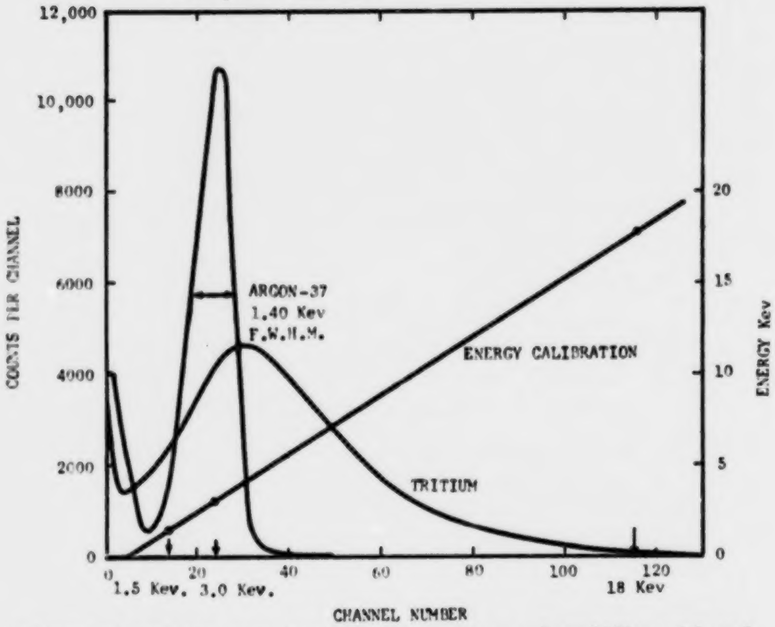
1. Sufficient counting time to achieve desired statistical precision.
2. Adequate shielding for stability of the background counting rate. Background radiation is usually measured only weekly (over weekends).
3. Stability and reliability of electronic components, with freedom from noise.
4. Maintenance of gas preparation and counter calibrations.
5. Minimum sample gas contamination.
6. Quality control checks of standards and background, with periodic interlaboratory comparison of samples and standards.

A review of the low-level counting systems of laboratories that participated in the IAEA/WMO world network of tritium measuring stations is found in Cameron [39]. An extensive literature review on sample preparation and counting up to 1967 is also included. An excellent review of low-level counting methods is given by Oeschger [40]. The several components contributing to the background radiation measured in counters are discussed and techniques for achieving "ultimate" background are evaluated. In recent work by Oeschger's group at Bern, significant advances have been made in low-level counting [41,42]. Geyh [11] presents methods based on practical experience for designing and constructing different types of detectors for low-background measurements.

For low-background counting, anticoincidence circuits and massive shielding are essential to minimize environmental ionizing radiation from cosmic rays, fallout radionuclides, ^{40}K and U and Th decay products. Since the background spectrum is different from the tritium spectrum, significant reduction of the background can be made in the proportional region by counting tritium using energy discrimination, with no appreciable loss in efficiency. For example, the

Figure 3

Tritium spectra with ^{37}Ar spectra superimposed. Gas mixture 3:1 HT:CH_4 at 4,000 torr.



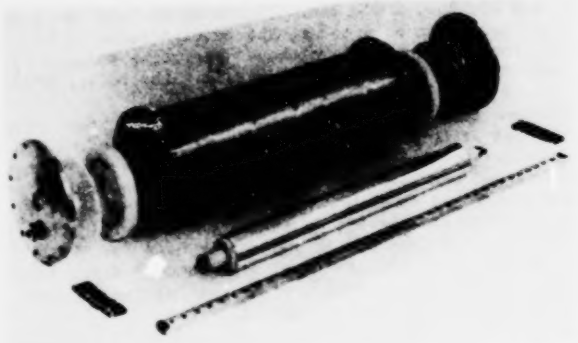
background counting rate in a typical detector and amplifier with and without energy discrimination is 1.5 and 5.5 cpm, respectively. The tritium spectrum measured with a detector in which a gas mixture of 3:1 HT:CH_4 is used in the proportional region is shown in Figure 3. The energy resolution of electrons is shown by the spectrum for ^{37}Ar X-rays counted in the same gas mixture.

It is apparent that in this system discriminators set to bracket the spectrum, between channels 5 and 120 for example, will measure essentially all of the tritium radioactivity. Since only about one-fourth of the total energy spectrum is needed in counting tritium, the ratio of background counts to tritium counts can be reduced significantly by judicious use of the "window" discriminators.

Figure 4 shows an Oeschger type of gas proportional detector [15], in which the total sample gas is placed inside the active volume, with guard gas in the outside or guard volume [43]. The separation between the guard-gas and sample-gas counting volumes is made by thin (3.5 to 7 mg/gm^2) aluminized mylar foil. Since the two regions are independent, a special pressure control device is necessary to fill and empty the chambers. In the normal Oeschger counter, the sample gas is used for guard as well and no separation is needed [44]. The guard is operated in anticoincidence to reduce the environmental background radiation. The thin window between

Figure 4

Photograph of membrane detector
from Schell 1969.



the guard and sample counters limits the background contribution from secondary electrons produced in the sample counter by energetic gamma or cosmic ray radiation interacting with the wall. This detector, of 1.14-liter volume, has been operated in the proportional region at 8,000 torr with a mixture of 3:1 HT:CH_4. At high pressure, it is possible to measure larger volumes of sample gas and maintain a low-background count rate. The practical limit of sensitivity is ca. 2 TR for 1,000 min counting time, requiring about 10 g water. The background is about 3 cpm inside the shielding consisting of 20 cm Pb, 10 cm paraffin, and 0.16 cm Cd.

At high pressures it is necessary to use metal lines. The hydrogen sample gas is condensed onto a small charcoal trap cooled to -196°C. The trap is warmed to ca. 20°C and the gas is compressed into the counter volume; methane is then added from a compressed tank at the counting pressure. Because of isotope effects on cooling and warming sample hydrogen during transfer, it is essential to warm the entire sample to approximately 20°C when filling the counter, to avoid fractionation. The use of sample hydrogen and tank methane gas fulfills the primary requirement of minimum gas handling to limit contamination.

Another detector [45,46] constructed of quartz, has background almost comparable to that of the Oeschger counter. In this counter, no special precautions are necessary in the filling procedure [46]. It may be possible to use these detectors at high pressures, though no low-level tritium counting has been reported in which a hydrogen-methane gas mixture was used and counting was done at high pressures.

The standard design detectors made from oxygen-free high conductivity copper (OFHC) in the USA, or "electrolytic grade" copper in Europe, can give low backgrounds. These are commercially available from several suppliers (LND, Reuter Stokes, Johnston Laboratories, Baltimore, MD). At the IAEA, the backgrounds of three 2-liter volume detectors inside a 20-cm iron shield are 1.1, 1.5, and 1.8 cpm. The same anticoincidence guard detector is used for the three detectors. The sample gas is ethane, prepared from tritium-free water and counted at 1,400 torr.

An external gamma-source position at a reproducible location inside the shield can be used to calibrate the voltage. A simple procedure may then be followed to check the purity of every gas filling. In a proportional counter, one of the upper channels, e.g., the "alpha" monitoring channel, is calibrated with a gamma source at a given count rate of about 2,000 cpm. If the count rate falls outside one standard deviation of a count, the voltage is adjusted accordingly. In routine work, the "plateau" operating voltage can be set accurately and is reproducible to within a few volts, usually after a few minutes' counting time.

One technique found to be useful for calibrating proportional detectors is that of using X-ray sources through a Be window. In this way the energy calibration can be made with different X-ray source energies. Looslie and Oeschger [47] use a thick wall plexiglas detector at 3,800 torr and calibrate with external X-ray sources. For high pressure detectors without a window, the conversion electrons and X-ray of ^{83m}Kr milked from a ^{83}Rb cow can be used for calibration. The half lives of ^{83m}Kr and ^{83}Rb are 1.83 hours and 83 days, respectively. Methane counting gas can be used as the "carrier" of the ^{83m}Kr tracer that is added to the counter [15].

ACKNOWLEDGMENTS

I would like to express my appreciation to Mr. Bryan Payne, Head, Isotope Hydrology Section, IAEA, Vienna, to Dr. Guy Sauzay and Dr. R. Brown, Isotope Hydrology Section, and to the members of the Isotope Hydrology Laboratory, Mr. Peter Schwarz, Mrs. Christine Schuesler, and Mrs. Mira Gattin, for their assistance.

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A CHROMATOGRAPHIC SYSTEM FOR THE ENRICHMENT AND ANALYSIS OF LOW-LEVEL TRITIUM SAMPLES

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An analytical procedure was needed to measure routinely tritium in the 5 to 150 TR range in a large number of water samples.

Florkowski, et al. [1] compared the results of analyses by 35 laboratories of tritium in natural waters in the range 10 to 250 TR and concluded that enrichment-gas counting was the more accurate method from 0 to 80 TR. Direct gas counting was more accurate for higher concentrations.

Electrolysis [2] and thermal diffusion [3] have been used to enrich tritium in water samples, but are lengthy procedures that require considerable attention from the analyst. Hoy [4], Perschke [5], and Akhtar [6] developed methods of tritium enrichment by chromatography.

Features and advantages of Hoy's gas-solid chromatograph include: relatively quick analysis of water samples containing less than 10 TR; self-elution that eliminates the need of an inert carrier gas; a completely sealed system that reduces tritium contamination; and simple collection based on pressure measurements (tritium evolves first). The technique is specific for tritium, because most other gases likely to be associated with the sample either do not react with the palladium column or are tightly bound. The columns are regenerated simply by simultaneously heating and evacuating the columns.

In this paper the automation and optimization of Hoy's analytical system are discussed.

EXPERIMENTAL

A tritium enrichment system was designed with a palladium column capacity of about 50 liters (STP) of hydrogen (∿40 ml water) per analysis preceded by a sample reduction furnace capable of producing up to 3 liters of hydrogen per minute from water. The assembly of the columns and magnesium furnace is shown in Figures 1 and 2. The water sample size was determined by the sensitivity objective of 5 TR (±10%) and operational parameters of the columns. Radioassay of the enriched fraction by proportional counting requires an enriched recovery equivalent to ∿1 cpm for a 5-TR water sample.

Previous experiments by Hoy [7] showed that a palladium gas chromatograph could reproducibly enrich hydrogen samples of varying concentrations (50 to 440,000 TR). The recovery was not dependent on concentration but was dependent on sample volume (56% recovery for a 50-ml sample; 80% for 10 ml). From a study of the effect of temperature on recovery, cooling the columns to -65°C would increase recovery

Figure 1

Assembly of the Chromatographic Columns

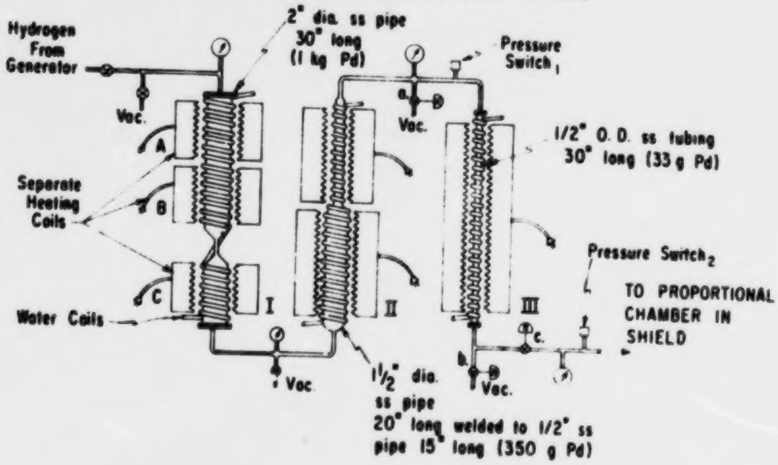
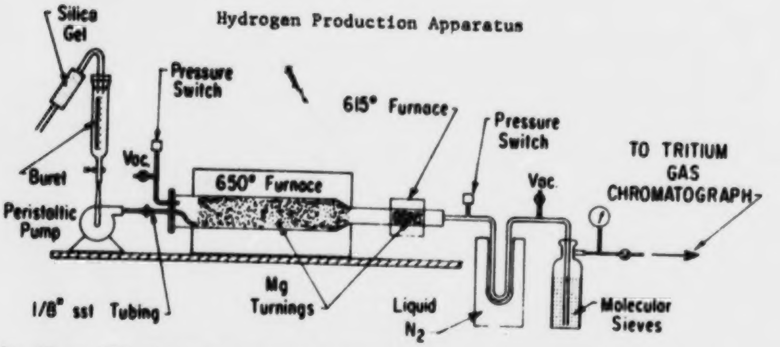

Hydrogen From Generator
2" dia. ss pipe 30" long (1 kg Pd)
Pressure Switch$_1$
Vac.
a.
b
Vac.
1/2" O. D. ss tubing 30" long (33 g Pd)
A
Separate Heating Coils
B
C
Water Coils
I
II
III
Pressure Switch$_2$
TO PROPORTIONAL CHAMBER IN SHIELD
c.
b.
Vac
Vac.
1 1/2" dia. ss pipe 20" long welded to 1/2" ss pipe 15" long (350 g Pd)

Figure 2

Hydrogen Production Apparatus



by 10 to 20% above that at room temperature, but would complicate the design and operation of the columns. The distribution of tritium in the enriched sample was determined as the tritium left the last column. Approximately 50% of the tritium was in the first 50 ml which decreased to ~0.6% in the 600 to 700 ml fraction. A volume of 630 ml was selected for counting and represented ~80% recovery with a negligible error if slightly different enriched volumes were collected.

Briefly, the enrichment scheme is as follows: The water sample is reacted with magnesium chips at 650°C in an enclosed furnance. The hydrogen generated sorbs directly onto an evacuated palladium column at ambient temperature. When a sufficient volume of water has reacted, the furnance is isolated from the palladium column. No buffer gas or carrier gas is used; movement of hydrogen through the column is maintained by the initial pressure differential (vacuum at exit end and hydrogen partial pressure at feed end) and the additional pressure created by zonal heating of the feed end of the column that causes the palladium to release sorbed hydrogen.

Tritium exhibits the highest partial pressure over palladium and diffuses through the column faster than either deuterium or protium. The first gas evolved, the tritium-rich fraction, is detected by a pressure increase in an evacuated proportional counter attached to the exit end of the column. After the tritium fraction is collected, the proportional counter is filled with propane gas and counted with a pulse-height analyzer to verify purity and electronic gain. The columns are regenerated by increasing the temperature of the entire system to 240°C and pumping out the residual hydrogen.

Details of the column and operational procedures are given in the Appendix.

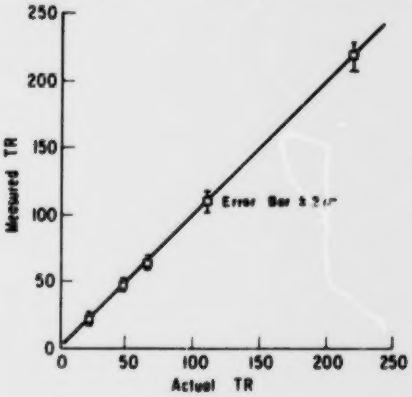
Certain practical points should be noted with respect to operating the column. Recovery and enrichment are low if any trace of oxygen remains in the system. All leaks must be eliminated and the columns stored under inert gases or vacuum when not in use. Chemical vapors that react with palladium or catalytically combine with hydrogen must be avoided, that is, halogens, sulfides, and oxygen. One and one-half hours are required for the tritium to evolve from the column, but other gases inert to the column pass through in a few minutes. Gases dissolved in water or those released from magnesium upon heating may be pumped off the exit end of the column without affecting the final result.

Results and Discussion

The conditions for the operation of the automated palladium-gas chromatograph are described in the Appendix. Average routine percen-

Figure 3

Column Analysis of Tritiated Water Standards



tage recovery, disregarding first sample after column inactivity, was 81.2 ± 1.6%. The first sample analyzed, after the columns were dormant for two days or more, averaged 9.5% below samples analyzed subsequently. Therefore, the first sample (a blank) analyzed on the column after a dormant period of two days or more is disregarded.

To investigate the analytical behavior of the system, a series of standards (22 to 210 TR) prepared from low-tritium, deep-well water were analyzed and results are shown in Figure 3. The precision of the system is also indicated by the fact that duplicate samples usually agree within the procedural statistics of ±6% (2σ).

A procedural contamination level of 4.2 TR was established by routinely analyzing deep-well water samples. It is uncertain whether this level of tritium is present in the well water or is the result of exchange with air moisture during sample handling and processing. Contamination between consecutive samples was investigated by analyzing a high activity sample (8,940 TR) and then immediately measuring a deep-well water sample in the system. An average residual tritium activity of 29 TR was found in the deep-well water sample. Approximately 0.34% ± 0.11 of the tritium from one sample contaminates the next one processed.

ACKNOWLEDGMENT

The information contained in this article was developed during the course of work under Contract AT(07-2)-1 with the U.S. Atomic Energy Commission.

APPENDIX

Chromatographic Columns

The palladium-column system has a capacity of about 50 liters of hydrogen gas. The assembly of the chromatographic columns is shown in Figure 1. Column I contains 1 kg of palladium mixed with 100 g of asbestos, and the mixture is supported in the column by stainless steel turnings. The column is wrapped with 1/4-inch-diameter copper water tubing for rapid cooling, three separate heating tapes (A, B, and C) for zonal heating, and insulation. The second and third columns contain lesser amounts of the palladium mixture (as indicated in Figure 1) and are wrapped with copper tubing, heating tapes, and insulation. All valves are bellows seal; connecting tubing is stainless. Swagelok* fittings are used for system couplings. The proportional counters, in a lead-water shield, are connected by stainless steel tubes to the exit of Column III.

Hydrogen Production Apparatus

A magnesium furnace capable of reducing a 40-ml water sample in less than 25 minutes and which could be evacuated to less than 50 µ was developed. The sample preparation system is shown in Figure 2.

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*Trademark of Crawford Fitting Company.

The 650°C furnace (containing ~170 g of magnesium turnings supported on either end by stainless steel turnings) is sufficient for the reduction of two 40-ml water samples. The small secondary furnace filled with 15 g of magnesium turnings is attached to the exit of the 650°C furnace. The furnace is positively fed by a small peristaltic pump at a rate of ~1.75 ml/min from a 50 ml buret. The intercoupled pressure switches are set such that the pump motor will shut off if the pressure reaches 10 inches of mercury (vacuum) and turn on when pressure drops below ~12 inches of mercury (vacuum). This technique allows uniform loading of the column and prevents pressure buildup in the glass manifold. The gas stream is passed through a liquid nitrogen trap, 200 cc of molecular sieves, and then to load Column I (Figure 1).

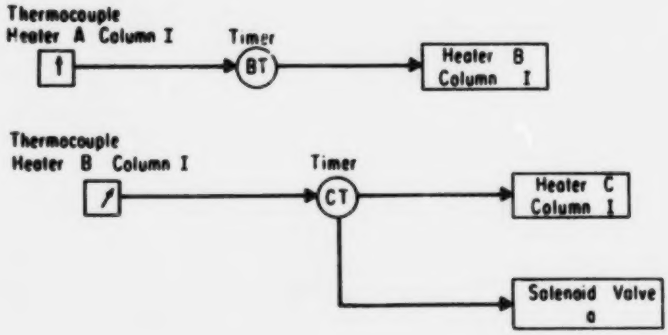
Operation

A buret is filled with the distilled water sample to be analyzed and attached to the inlet tube of the peristaltic pump. The prepared furnace (heated to 500°C overnight) and hydrogen purification apparatus are evacuated to 10 μm of Hg. Column I and sample preparation apparatus are isolated from vacuum and the sample is fed to the furnace at a rate of ~1.8 ml/min. Once the sample (40 ml) is reduced and load system pressure decreased to 20 inches of mercury (vacuum), Column I is isolated from the sample preparation apparatus. The elution sequencer is reset to automatic position, and elution of the tritium peak is initiated by turning on heater A.

When heater A reaches 240°C (~3 min), timer BT (Figure 4) is started, and at the end of 7 minutes heater B is turned on. When heater B reaches 240°C, timer CT (Figure 4) is started, and at the end of 7 minutes heater C is turned on and automatic valve "a" (Figure 1) is closed. Approximately 20 minutes after heater A is turned on, a positive pressure is shown on the gage between Columns I and II.

Figure 4

Elution Sequencer



Forty-two minutes after heater A is turned on, the pressure between Columns II and III has increased from 30 inches to 25 inches of mercury (vacuum), and automatic valve "b" is closed leaving the exit of Column III open to the proportional chamber. Nine minutes after valve "b" is closed, a pressure is registered at the exit of Column III, and 3 to 5 minutes later pressure switch 2 closes off the proportional chamber at 22 psi and reopens automatic valve "b" to vacuum. Total time of elution sequence is 55 to 60 minutes.

The columns are then opened to the vacuum manifold, elution sequencer is set to manual, heaters to Columns II and III are turned on, and system is pumped down to 200 μm of Hg (~1.5 hours). After cooling to ambient temperature by passing air, water, then air again through the cooling coils, the pressure of the system is reduced to 20 to 30 μ. At this point a second sample may be analyzed.

The sample in the proportional chamber is increased to 30 psi with propane, high voltage applied, and counted with a pulse-height analyzer.

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REVIEW OF TRITIUM MONITORING DEVICES

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Tritium, the radioactive isotope of hydrogen, is formed abundantly in various nuclear reactors. A possible rupture or leak in water lines may result in a radiation health hazard by spreading significant quantities of tritiated water into the atmosphere. Accidents and poor handling procedures in industry and various research laboratories utilizing the isotope in high activities may also result in acute or chronic exposures, especially if tritiated water is involved. Physically, it may appear that tritium is harmless due to a beta spectrum characterized by a maximum energy of 18 keV and an average energy of only 5.5 keV. The average beta range in water is only a few microns and can hardly be considered hazardous as an external source of radiation. However, tritium in the oxide state is readily absorbed by the lungs and skin and is assimilated into body fluids. The relative hazards presented by environments contaminated with HTO vapor in contrast with tritiated elemental hydrogen is demonstrated by the maximum permissible concentrations (MPC) of 5×10^{-6} μCi/ml and 2×10^{-3} μCi/ml, respectively.

The need for adequate monitoring techniques has led to the investigation of many possible monitoring systems. The difficulties associated with the detection of low energy radiation have necessarily complicated basic detection devices, and individual specifications are, therefore, often necessary for each particular monitoring requirement. Particular applications require consideration of factors such as type of media to be monitored, desired sensitivity, possible effects of the surrounding environment, and a host of other equally important parameters before a successful tritium monitor can be designed, constructed, and proven useful. In short, almost any successful monitoring system can be regarded as an accomplishment.

Practical monitors can be separated into two groups. One group consists of all monitors that render constant or immediate indications of activities within the monitored media. The second includes all tritium collection systems that utilize water vapor collection traps for subsequent analysis of tritium. The immediate readout feature of the first group and high sensitivity of the second may make the use of either one more attractive than the other in a given situation, or may require the use of both.

Constant Readout Monitors

The basic components of the first group, constant readout monitors, are the detector and its associated electronics, readout and, if needed, a system that constantly exchanges the monitored medium. The heart of the system is the detector, usually consisting of an ionization chamber, proportional counter, or scintillator-photomultiplier (PM) tube combination. Since the ionizations chamber collects the ions produced by ionizing radiation, a current amplifier comprises the associated electronics. In contrast, the proportional counter and PM tube are normally pulse devices and therefore require pulse-handling electronics.

The readouts most often used for the various systems are meters that may be calibrated with either counts per minute, units of specific activity, increments of MPC, or other values that may be applicable. These units are all interrelated and may be converted using proper calibration constants. A second form of readout is the scaler that is used with pulsed systems. This readout is not constant, but the integration period can be made short and repetitive, thereby simulating a constant readout and simultaneously increasing sensitivity.

With three detection devices available, one may infer that each has its merits and, similarly, its limitations. A brief examination of features peculiar to the devices substantiates this inference.

The Ionization Chamber

The current available from the ionization chamber is simply the rate of ion collection from inside the chamber. These ions can be caused by any type of ionizing radiation of any energy; therefore, energy discrimination and the ability to partially distinquish the tritium component from components due to other radiations are impossible. Also, the current is usually extremely minute. A simple calculation indicates a maximum current of 4.8 X 10^{-15} A/l·MPC. The sensitivity of an ionization chamber is therefore dependent upon chamber volume and amplifier response. With currents this low, a number of seemingly insignificant factors may cause significant errors. Waters [1] reports these factors to be cigarette smoke, aerosols and ions, moisture and condensation, ambient gamma-ray field changes, radon, clogged filters, fission product gases, absorption and memory effects, and instrument calibration factors. Thus, with so many precautions necessary for its use, a practical ionization monitor usually employs a filter and ion trap at the input. The use of gamma compensation, by means of a similar but closed chamber that negates response to gamma, is also beneficial.

As impractical as it may appear, the ionization chamber is the most popular device for general tritium detection in air. The construction is relatively simple and the measurement of currents has been easier than the measurement of pulses. The chamber came into use earlier than the other detectors and has therefore become a recognized standard. It is also accurate if used judiciously.

As stated earlier the maximum current possible is only 4.8 X 10^{-15} A for each MPC contained in one liter. The Kanne chamber (Figure 1) monitor which is used at the Savannah River Plant illustrates the necessity of large chambers if high sensitivity is to be achieved. According to Morris [2] of this installation, two sizes of chambers, 51.5 liters and 18.5 liters, are used routinely. The absence of a gamma compensation device requires installation of the chamber in a low background environment, but remote areas can be monitored by use of hoses. The sensitivities for the 51.5 and 18.5 liter chambers are 2 and 8 pCi/ml, respectively.

Figure 1

Diagram of Kanne Chamber

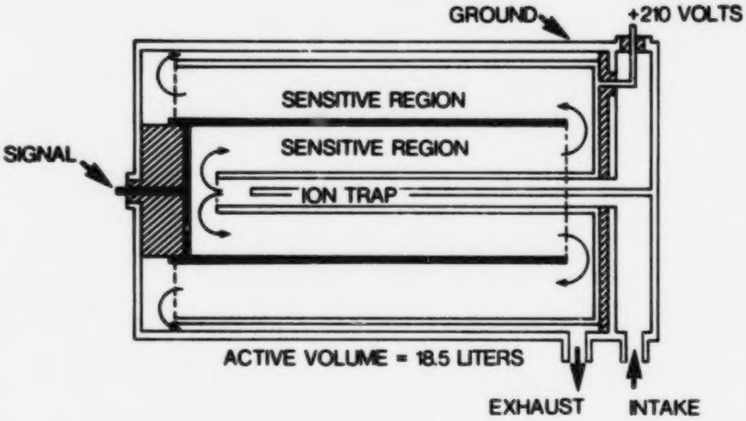
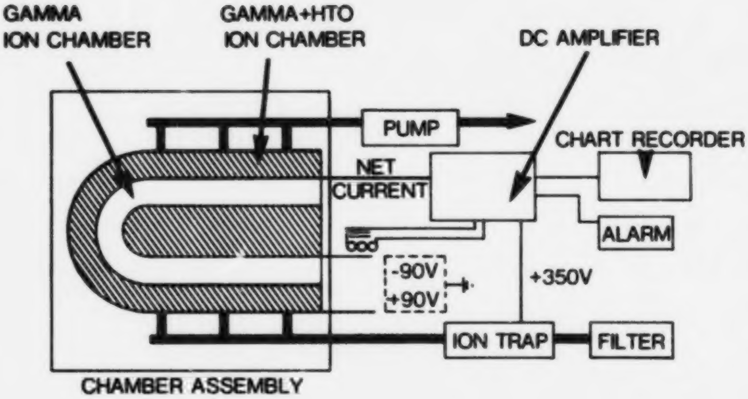


Figure 2

Osborne Tritium Monitor



Various designs for gamma-compensated ionization chambers can be found. Anthony [3] describes a compensated system that uses a rectangular-shaped sensitive chamber and a similar but closed chamber adjacently mounted for compensation. The device has overall dimensions of 5 X 10 X 16 inches and a reported sensitivity of 40 pCi/ml.

A similar detection device is the Pittendrigh and Vousden [4] chamber, consisting of two identical half-cylinder chambers coupled across the plane surface. These also are constructed so that one chamber is closed and serves as a compensation device.

A system with gamma compensation of a different design is that of Osborne and Cowper [5]. Because the spatial separation of chambers in previous designs has made them dependent on gamma field direction, the compensating chamber is placed inside the active chamber (Figure 2) so that geometrical symmetry provides equal response in essentially all directions. This arrangement is used in an area monitor of 40-liter capacity and two smaller portable versions of 1.2-liter and 0.3-liter capacities. The most sensitive scale for the 40-liter chamber is 0 to 1.5 MPC, or 0 to 7.5 pCi/ml. For the smaller chambers, a portable vibrating reed electrometer makes possible the corresponding scales of 0 to 10 MPC and 0 to 45 MPC for the 1.2-liter and 0.3-liter models, respectively. An interesting feature, however, is the inevitable change in compensation due to different ion collecting efficiencies for the electrodes.

The Proportional Counter

The proportional counter is similar in principle to the ionization chamber, but differences in design and bias and the requirement for a particular counting gas change the chamber into a pulse device. The proportional relationship between the pulse height and the number of ions produced per each absorbed ionizing particle allows the use of energy discrimination circuitry. This capability is significant in reducing monitor response to radiation other than the tritium beta. The necessity of keeping the counting gas intact require premixing with the monitored gas in specified proportions. This process inhibits general use of the counter, and continuous use at high sampling rates can be uneconomical. The sensitivities obtained with such counter, however, can be extremely high. An alternative method of designing a counter which is conservative in gas use is the thin window proportional counter. In this case, the tritium beta must penetrate the thin film that forms part of the counter's walls. Procurement of film which is thin enough, however, is a considerable hindrance.

The use of proportional counters for tritium monitoring was initiated by Driver [6], who was easily able to detect 20 pCi/ml with a counter volume of only 43 ml. This sensitivity was later increased by Ehret [7] to 0.1 pCi/ml or 0.02 MPC using counters with less than one-liter volume. To obtain these sensitivities, commercial counters were used which employed complex electrode arrangements (Figure 3) which, when used with appropriate electronics, made energy discrimination, anticoincidence and gamma compensation possible. These counters, however, are of the flow-through type which may make them impractical for general monitoring.

Figure 3

Multi-Electrode Proportional Counter

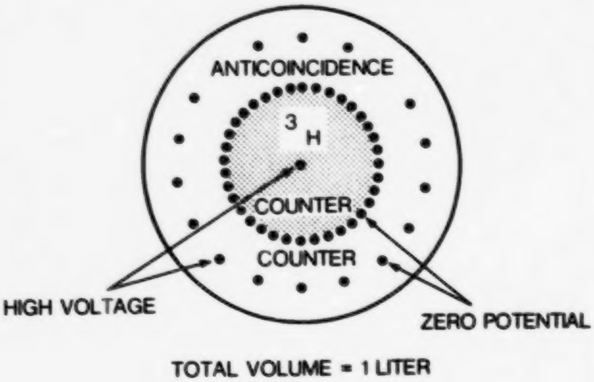
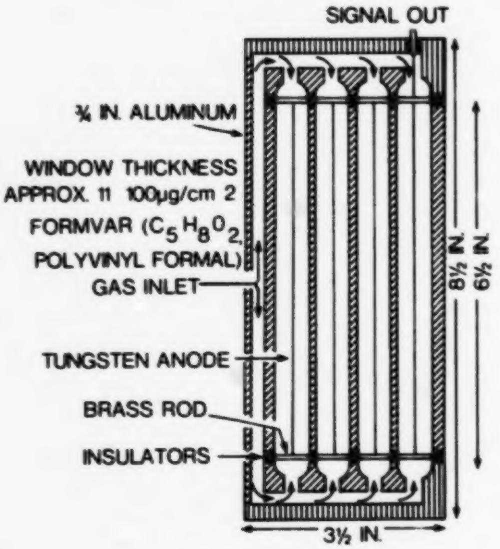


Figure 4

Internal View of Thin Window Proportional Counter



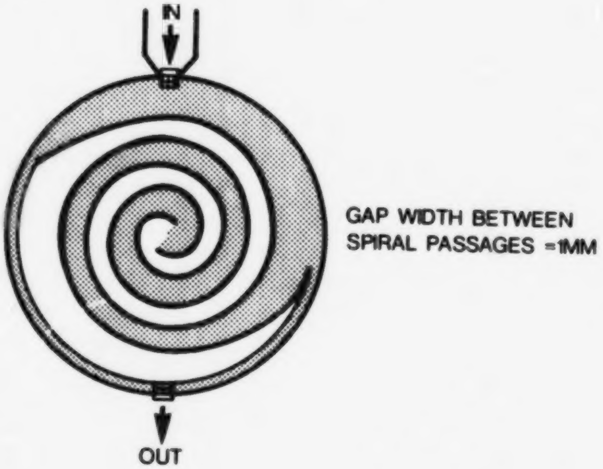
A thin window proportional counter designed for general monitoring has been described by Block and associates [8,9]. This monitor, like the ionization chambers, employs two identical counters (Figure 4), one of which is used for gamma compensation. Because the thin window over the compensating counter is opaque only to tritium beta, compensation occurs also for high energy beta and alpha. The response for this system is 577 cpm/MPC.

The Scintillator-PM Tube

The third type of detector used in tritium monitors is the scintillator-PM tube detector and it differs greatly from the previous detectors. In this detector, absorbed ionizing radiation causes the scintillator to emit photons of light. If detected by the PM tube, a pulse of current is generated. The proportional relationship applies also for this detector and use of energy discrimination circuitry is usually employed. An outstanding feature of this detector over the other two is its ability to monitor liquids directly. The mechanics associated with this device which determine its practicality are too complicated to briefly state but are essentially identical to those associated with the liquid scintillation counter. The use of scintillators for tritium monitors has been necessarily delayed in recent years, but compact electronics, high quality scintillators and PM tubes have now brought attention to their use and promise.

Figure 5

Spiral Scintillator Detector

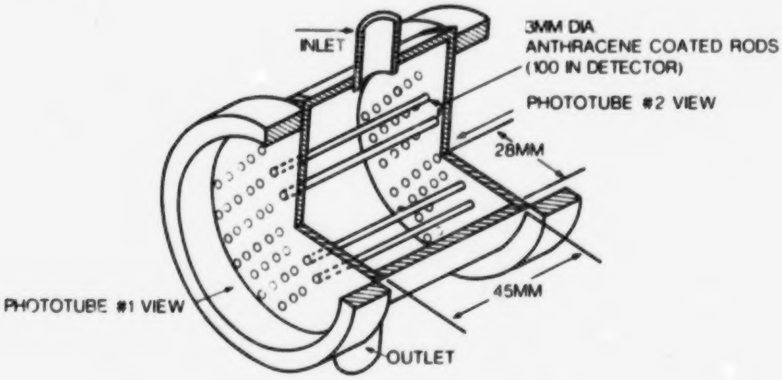


Because of the availability and convenience offered by plastic scintillator, its use in tritium detectors has been explored more than any other scintillator. With the detector geometry shown in Figure 5, Sannes and Banville [10] reported a sensitivity of 0.2 MPC. The detector consisted of two strips of plastic scintillator rolled together to form a double spiral. By making use of the energy discrimination circuit in the electronics, gamma compensation was performed. The entire system was designed with batteries as a source of power and protability was therefore accomplished.

A detector using 40 parallel 2-inch-square sheets of plastic scintillator was constructed at the Eastern Environmental Radiation Laboratory (EERL). It was tested with electronics equivalent to those in liquid scintillators. The sensitivity also was found to be 0.2 MPC but required use of lead to shield the detector from local gamma background. Static electricity was observed at times and memory effects from tritiated water were judged to be too severe. The plate detector was temporarily abandoned and the design of Figure 6 was investigated [11]. This detector, using a thin layer of anthracene coating on the rods, proved to exhibit a sensitivity of 0.2 MPC also. The use of anthracene greatly reduced background sensitivity, and no shielding was required for this model in the local environment. The memory effects were also greatly reduced and the design allowed monitoring of water and filtered urine with a sensitivity of 10 cpm for each μCi/l. Prolonged use of this detector proved, however, that anthracene did not make a suitable scintillator for a practical detector because of quenching, presumably from oxidation.

Figure 6

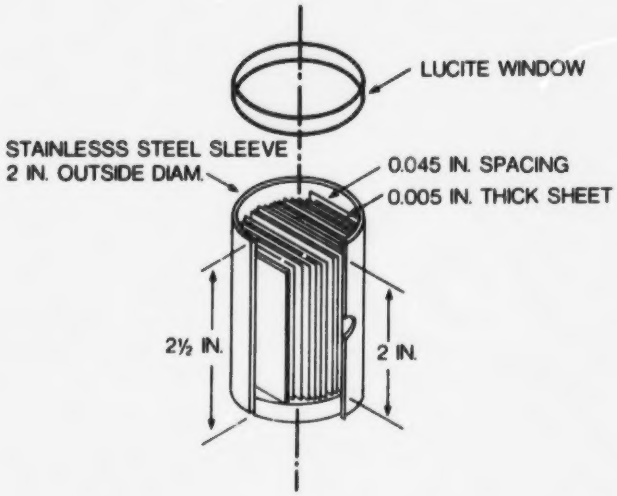
EERL Rod Detector



Other parallel-plate detectors have been investigated by Prevo and Santomassimo [12] (Figure 7) and Osborne [13]. The latter's detector, consisting of sixty 5 cm X 5 cm plates, rendered impressive results. With this detector, a response of 24 cpm for 1 μCi/l of water was observed. The same detector also exhibited the extremely high response of 150 cpm pCi/ml, or rather, 750 cpm/MPC for tritium in air.

Figure 7

Parallel Plate Scintillator



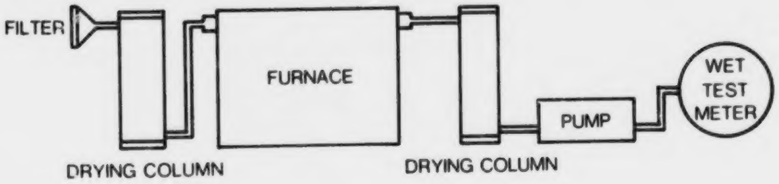
Two other detection systems that should be mentioned are the tritium-in-air detector of Prevo and Santomassimo [14] and the automatic tritium-in-urine analyzer that was designed by Osborne [15]. The former system uses a single crystal of CaF_2(Eu), a nonhydroscopic inorganic scintillator which does not suffer from memory effects commonly observed with other scintillators. The latter system automatically mixes aliquots of urine with liquid scintillator and analyzes the solution for tritium.

Tritium Collection Systems

The second group of monitors which use water vapor collection traps may consist simply of an air pump, air volume metering device, and a single trap. A more complex system would be required, however, if accurate determinations are to be made for air containing different forms of tritium. The sensitivity of these systems depends primarily

Figure 8

Combustion Train



upon the volume of air sampled and the type of trap used. The decision of what trap to use is determined by weighing the convenience, sensitivity, and accuracy afforded by the various traps. Air that is pumped through a desiccant such as Drierite (W. A. Hammond Drierite Company, Xenia, OH), silica gel and molecular sieves will lose almost quantitatively its water vapor to the desiccant as long as saturation is not approached. The vapor is then retrieved by distillation. If a lower sensitivity is tolerable, the vapor can also be retrieved by mixing the desiccant with distilled water and letting the solution sit until displacement has occurred.

Traps often used, other than desiccants, are cooled condensation traps and water bubblers. The cooled condensation trap yields high sensitivity since aliquots of the condensed vapor can be obtained without dilution. However, care must be exercised to ensure complete collection. If incomplete but constant collection can be tolerated, the water bubbler may often prove to be satisfactory. Investigations [16,17,18] have shown that collection efficiencies above 95% can be obtained with relatively small volumes of water. The bubbler does necessitate dilution, but a lower sensitivity may not be considered a hindrance due to the convenience afforded.

An example of this group that has been used successfully at EERL is shown in Figure 8. Sequential components in this particular system are: (1) particulate filter, (2) Drierite canister for removing water vapor, (3) combustion chamber for converting hydrocarbons to water, (4) Drierite canister, (5) air volume meter, and (6) air pump. The filter and Drierite canisters are analyzed in the lab after sample collection for accurate specific activity determinations.

Conclusion

The selection of the various units, μCi/ml, cpm/(μCi/ml), cpm/MPC, and MPC, used to indicate a system's ability to detect tritium was not deliberate. These were the units chosen by the various authors. Sensitivity should mean the threshold of activity at which an instrument begins giving a meaningful measurement. Unless the conditions are stated in which the measurement takes place, it must be assumed that the measurement is made under ideal conditions, for obviously, in high gamma fields, any instrument's "good sensitivity" can be destroyed. The system's response such as cpm/MPC, however, remains essentially constant and is therefore more descriptive.

In closing, the foregoing systems have illustrated only partially the various systems and techniques that can be used for tritium monitoring, and a sincere apology is due to other authors of meaningful contributions. Hopefully, one may infer that any situation may require its own peculiar system of monitoring. The parameters that affect the final activity determinations must be taken into account, and complete knowledge of a system's limitations is of extreme importance.

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MEASURING TRITIUM WITH LIQUID SCINTILLATOR SYSTEMS

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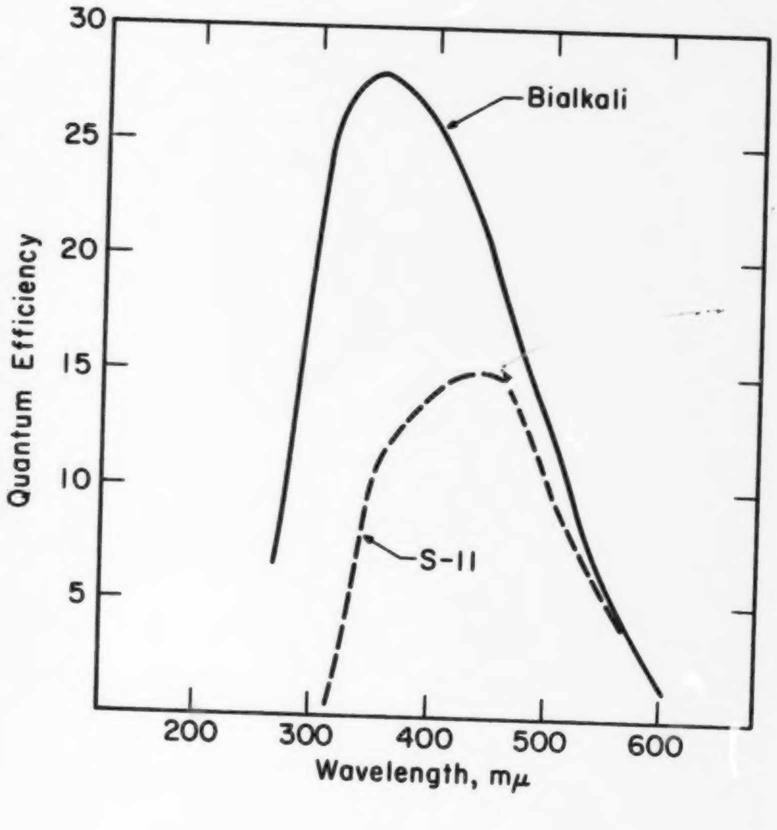
Liquid scintillation counting has been shown [1,2] to be an accepted, and often the only, way to measure the amount of tritium present in many different types of samples. Liquid scintillation counting is based upon the fact that during the stopping of a radioactive emission (alpha particle, beta particle, gamma ray, etc.) in an organic solution, excited molecules are produced [3]. Upon the spontaneous return of these excited molecules to their ground state, photons are emitted. Each excited molecule has the possibility of emitting only one photon. The fraction of the excited molecules which do emit photons, as opposed to radiationless processes, is known as the quantum yield. Most common scintillator solutes have quantum yields near 90% [4]. The number of photons emitted is proportional to the number of excited molecules produced during the stopping of the radioactive emission. Many excited molecules are formed even for the stopping of very low energy beta particles, such as those emitted from a tritium decay. The burst of photons is detected with a very sensitive multiplier phototube (MPT) which amplifies the signal and generates a voltage pulse proportional to the number of photons which fell upon the photocathode of the MPT. The output of the MPT can be used to drive a rate meter (to measure count rates) or analyzed by a pulse height analyzer (to sort and record events proportional to the intensity of each event).

The liquid scintillator solution consists of three main parts. The bulk of the solution is the solvent, usually an alkyl benzene (i.e., toluene) or dioxane. To the solvent an organic compound (or compounds) is added which is an efficient photon emitter and which emits photons in the wavelength region which is easily measured by the MPT. And finally the liquid scintillator solution contains the sample; although there are some uses where the excitations are generated by an external sample. The most troublesome problems encountered with liquid scintillation counting involve those associated with the introduction of the sample into the scintillator solution in a homogeneous (or near homogeneous) form, without causing a drastic reduction in photon emission due to so called quenching processes.

In the liquid scintillation counting process, a pulse (or count) is recorded for each radioactive emission which is sufficiently energetic to produce a detectable number of photons. Some events, which for various reasons (low energy, quenching, etc.) do not produce a measurable number of photons, will not register a count in the liquid scintillation counter. Thus, the counting efficiency for detection of a radioactive material with many low energy particles will be less than 100% [5]. For modern liquid scintillation counters and non-quenching organic samples containing tritium, the tritium

Figure 1



counting efficiency will be nearly 65% depending upon the low energy bias necessary to reduce the electronic background.

The main advantage obtained from the use of liquid scintillation counting as opposed to other conventional counting methods has to do with the fact that the sample containing the radioactivity is in intimate contact with and, in most cases, dissolved in the detector. This produces the ideal 4π geometry. A limiting factor in the liquid scintillation counting system is the MPT. Even the best MPTs have photocathode efficiencies for conversion of photons into electrons of only 28%. The MPT efficiency is also dependent upon the wavelength of the photons, see Figure 1. The efficiencies are even less for wavelengths greater and less than the peak response.

The photocathode efficiency will limit the efficiency for counting radioactive emissions. The detection efficiency for electrons of different energy is shown in Table 1 for single MPT detector and for two MPTs in coincidence. The values are based on the scintillation yield of 0.04 from Lipsky [6]. At low electron energy the efficiency in the coincidence MPT system deviates markedly from the 100% efficiency for higher energy electrons. The counting efficiency of less than 100% for a beta-emitting nuclide, e.g., tritium, results from the fact that a certain fraction of emitted electrons (beta particles) does not produce enough photons to be detected by the MPT (i.e., a continuum of beta particles is emitted).

Table 1

Average Number of Photons, Photoelectrons and Expected Counting Efficiency as the Function of the Energy of the Electron Stopped in a Liquid Scintillation Solution

Electron energy keV	Average number of Photons	Average number of photoelectrons at photocathode efficiency of		Counting efficiency[a] %, coincidence counter with two MPTs with efficiency of	
		15%	28%	15%	28%
1,000	12,500	1,875	3,500	100	100
500	6,250	938	1,750	100	100
158	1,980	297	555	100	100
50	625	94	175	100	100
5	63	9	18	99.6	100
1	13	2(0.86)[b]	3.5(0.97)[b]	37[c]	77[d]
0.1	1.3	0.2(0.15)[b]	0.4(0.27)[b]	0	0

[a] Calculated for counting system which does not have pulse summation: efficiencies will be a little higher for low-energy electrons with pulse summation, but those events which do not produce enough light to give at least one photoelectron in each MPT will not be counted in either system.

[b] This fraction of the time the number of expected photoelectrons will not produce a measurable pulse: i.e., for 2 photoelectrons, 14% of the time no measurable pulse will be produced. Also this means that the production of only enough photons to give on the average 0.4 photoelectron will produce a measurable pulse 27% of the time.

[c] This efficiency is calculated from the probability of coincidence ($Y = 1-2^{(1-R)}$ where R is the average number of photoelectrons) and the square of the probability of a measurable pulse from an MPT: $0.50 \times (0.86)^2 = 0.37$

[d] Using the immediate above example: $0.82 \times (0.97)^2 = 0.77$

Solvents and Solutes

The solvent serves two functions: (a) the primary excited molecules formed during the stopping of the radiation are excited solvent molecules, and (b) the solvent facilitates the dissolution of the sample in the liquid scintillator solution.

The production of excited solvent molecules is most efficient for those types of organic compounds which have electronic systems which include easily excited non-bonding electrons (called π electrons) with low lying energy levels near 2 to 3 eV above the ground state [7]. Benzene, the alkyl benzenes and dioxane all have these π electron systems. Solvents such as saturated hydrocarbons (i.e., n-alkanes, cycloalkanes, etc.) do not have π electron systems and are not efficient solvents for liquid scintillator solutions.

The choice of solvent will sometimes be determined by the nature of the sample: organic soluble vs. water soluble. Benzene

Table 2
Some Fluorescence Properties of Some Solvents, Primary Solutes and Secondary Solutes Used in Liquid Scintillation Counting

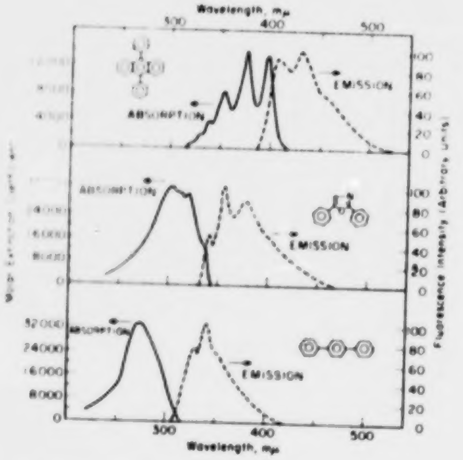
Compound	λ_{fl}, mμ[a]	Quantum Efficiency[b]	Decay time, 10^{-9} sec	Relative Scintillation Yield[c]
Benzene	280	0.07	29	85
Toluene	285	0.14	34	100
p-Xylene	290	0.30	30	110
Dioxane	250	0.03	2.2	65
Cyclohexane	200	0.004	--	20
p-Terphenyl	342	0.84	1.0	93
PPO	370	0.90	1.4	100
PBD	367	0.80	1.5	110
Naphthalene	334	0.19	500	---
DM-POPOP	427	0.85	1.5	---
BBOT	440	0.60	1.1	---

[a] Average wavelength of the fluorescence emission.
[b] Fraction of excited molecules which de-activate via fluorescence.
[c] Relative to deaerated solution of 3 g/l of PPO in toluene equal to 100. Values not given for the secondary solutes because they are usually used in the presence of a primary solute, i.e., PPO.

and alkyl benzenes are used for samples which are directly soluble in organic systems. Aqueous samples are most often dissolved in dioxane solvent solutions or suspended in alkyl benzenes by use of emulsifiers or gelling agents.

The solute is selected by optimizing several parameters. The solute should emit photons in the wavelength region of maximum quantum efficiency of the MPT. It is desirable to have a solute with a short decay time (1 to 2 X 10^{-9} sec) [4]. The shorter the decay time the less the probability of quenchers (e.g., oxygen) interacting with the excited molecule before it emits the photon. Solutes with a high quantum efficiency are preferred. (Quantum efficiency is the fraction of excited molecules which emit photons via Fluorescence.) A small overlap of the absorption and emission spectra is desirable to minimize re-absorption of emitted photons by the solute itself. This is very important for concentrated solutions and large volume scintillator solutions. Table 2 lists some data for several of the most commonly used scintillator solvents and solutes. Figure 2 shows the variation of spectral overlap with some commonly used scintillator solutes.

Figure 2



The solute must also be chemically inert to material introduced into the solution. Its solubility must not be affected by the introduction of the sample into the solution. It should have a high efficiency for energy transfer at moderate concentration. It should also be stable toward radiation damage (initiated by the radioactivity of the sample).

Sample Types

Several different types of samples are encountered when experiments are carried out to measure tritium with the liquid scintillation counter. In recent practice many samples containing organic tritium have been combusted in "oxidizers" and the tritium measured as THO. Commercial instruments are available which efficiently convert organic matter into CO_2 and H_2O by combustion in an O_2 atmosphere. The tritiated water is incorporated into liquid scintillator solutions by several different techniques, see Table 3.

Table 3

Some Techniques for Including Tritiated Water into Liquid Scintillator Solutions

Agent	Sample Form	Comments
Dioxane as the solvent	Homogeneous solution	a) High freezing point leads to difficulty in cooled counters.
		b) High water content with complete solubility.
		c) Dioxane decomposition leads to quenching.
		d) Chemiluminescence is present.
Ethanol-toluene	Homogeneous solution	a) Limited water solubility.
		b) Some phase separation and wall plating in cooled counters.
		c) Ethanol quenching - although less than dioxane quenching.
Emulsifiers [11,12] and suspension and gel	Heterogeneous solutions, droplet size is very critical	a) Varying physical state with water content.
		b) Wide range of water content with good counting efficiencies.
		c) True solution at low water concentration with emulsifiers.
		d) In heterogeneous solutions, it is difficult to determine quench corrections.

Of course there are also many techniques used for incorporation of the organic compounds directly into the scintillator solution. The choice will vary with the characteristics of the sample. Polymeric material often has to be chemically degraded before it can be dissolved in the scintillator solutions. Other organic materials may have to be treated (chemically and physically) to remove color or quenchers from the samples.

Sample Forms for Counting

Some of the counting techniques employed for measurement of tritium-containing samples in a liquid scintillation counter are listed in Table 4. In some cases the sample and scintillator solution form a homogeneous system, whereas other methods employ heterogeneous sample and scintillator systems.

Table 4

Some Counting Techniques for Measuring Tritium-Containing Samples

Technique	Examples	Refs.
1) Conversion of organic compounds by chemical reaction	Degradation of polymeric materials	13a
	Use of solublizers	13b
	Oxidation: wet and O_2 flame	13c, 14a 14b
2) Flow cells Heterogeneous	Flow of sample over scintillator beads or crystals	13d
Homogeneous	Pre-mix samples and scintillator solution: allow mixture to flow through a counter at a known rate and measure either total activity or changes in activity level	13d
3) Solid supports	Filter paper, glass fibers	13e, 14c
	Gels	14d
	Emulsions	13f, 12, 15, 16
4) Exchange reactions	Catalysed isotopic exchange of tritium from pure water into aromatic hydrocarbons	14e, 17
5) Catalytic hydrogenation	Conversion of sample to H_2(TH) and addition to unsaturated hydrocarbons which are soluble in liquid scintillator solutions or synthesis of the aromatic hydrocarbon solvent: i.e., benzene	18, 19

Quenching

Of prime importance in liquid scintillation counting is the possible presence of different amounts of "quenching" in supposedly identical samples [8]. Quenching is defined as any of those mechanisms which lead to a decrease in the number of photons emitted by a scintil-

Table 5

Some Methods for Measuring Sample Quenching Effect Upon Counting Efficiency in Liquid Scintillator Solutions

Method	Comments	Refs
Internal standard	Addition of small amount of high specific activity of isotope being assayed. For best results added material should be same form as measured sample. Sample cannot be recounted.	20, 21
Sample channels ratio	Measure ratio of the counts in two selected channels. Ratio will vary with amount of quenching. Not useful for low counting rate samples. Difficult to apply for dual labeled samples. Gives very accurate results.	22, 23, 24
External standard	Use gamma ray source to produce Compton scattered electrons which excite the liquid scintillator solution.	
count rate	Count rate changes with amount of quenching. Very critical of position of gamma ray source and changes in the electron density of solution and vial.	25, 26
channel ratio	Measure ratio of counts in two selected channels. This method is not so critically affected by the gamma ray source position and changes in electron density.	27, 28
Compton edge	Measure to pulse height value of the Compton edge as the function of counting efficiency and quenching. Requires a multi-channel analyser. Difficult with very high amounts of quench and not compatible with many commercial counters.	29, 30, 31
	In these methods the samples can be recounted. High activity gamma sources can be used to reduce counting time without fear of contamination of the sample.	
Quenching curves		
internal	Measure the counting rate with addition of increasing amounts of quenchers and extrapolate to zero quench. Need to make several additions and recounts of the sample. Sample cannot be recounted.	32, 33
external	Measure the counting rate with varying standard absorbing filters between the sample and the PMT. Extrapolate to zero absorbency. Since external, the sample can be recounted. Need filters of known absorbency, but they can be used for all samples.	14
Efficiency stick	Measure count rate of sample deposited upon a "stick" which is inserted into the solution. Activity must be fixed so as not to contaminate sample. Not useful for high counting rate samples. Requires corrections for half-life decay.	34, 35
Extinction coefficient	Measure count rate variation with quench as monitored by the extinction coefficient of the solution. Requires second measurement in an absorbence spectrophotometer.	36, 37, 38

lator solution. There are two basic types of quenching; chemical and color. Chemical quenching involves the action of foreign materials upon the excited molecules: either solvent or solute molecules. Energy is removed from the excited molecules by a process which does not involve the emission of photons. Color quenching involves the re-absorption of emitted photons by foreign material which may be present in the liquid scintillator solution or sample. The re-absorbed photons will produce excited molecules which de-activate by non-radiative processes.

The overall effect of these processes is the decrease in the average number of photons emitted per unit energy of exciting particle stopped in the solution. It is important to keep in mind that a 50% quenching of photon yield does not mean a 50% decrease in counting efficiency. Indeed the same amount of quenching will produce a greater change in counting efficiency for tritium than for ^{14}C. A 50% decrease in photon yield for excitation by an alpha emitter still gives a 100% counting efficiency for the alpha particles [9].

Several techniques used for quenching corrections are listed in Table 5. For homogeneous samples the internal standard technique is probably the most accurate but this has several drawbacks: the sample cannot be re-counted; opening the sample increases the chance of contamination; the standard has to be introduced in the same form as the sample, etc. The use of an external standard is the most widely practiced technique. Techniques are used which relate the amount of quenching either to the total count rate of the external standard or to the ratio of the counts in two specially selected voltage windows (channels). Typical plots for these two methods for tritium quenched samples are shown in Figures 3 and 4. The channels ratio method is more accurate since it is not affected as much by the positioning of the gamma ray source or the electron density of the sample.

Figure 3

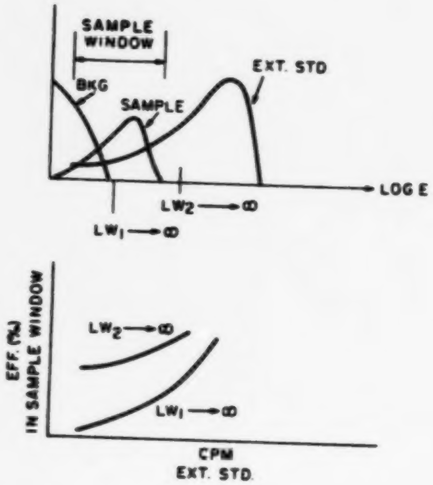
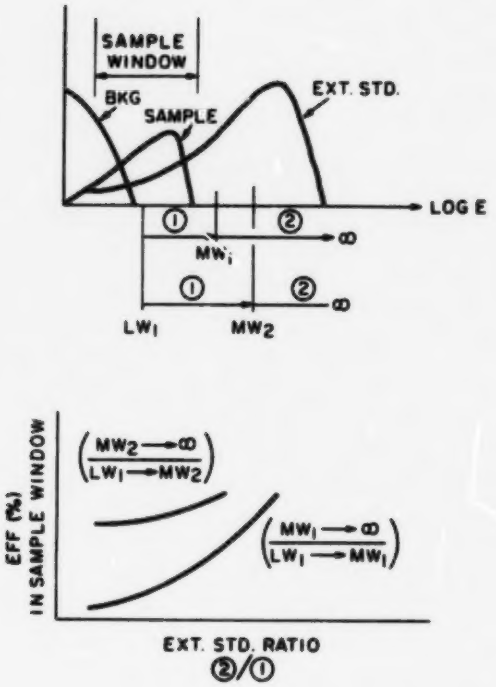


Figure 4



Background

Many times, experimenters have concluded that one background can be used for all samples. For high to moderate counting rate samples this assumption probably does not lead to any serious errors in determination of the count rate. However, it should be remembered, with modern liquid scintillation counters with their short resolving times (10 to 70 X 10^{-9} sec), that the background is mostly due to radioactive emissions exciting the scintillator solutions. These emissions are from cosmic radiation and natural and laboratory radioactive sources. The excitations produced by these sources are subject to the same quenching processes as are excitations produced by emissions from the sample. Therefore, quenching can also lead to a decrease in the background rate. (In some special cases where high-energy background radiations are present, quenching may produce an increase in background rate in a window selected for counting low energy emissions.) Thus, it is very important that a background rate be determined as a function of the quench in the same manner as the counting efficiency. Some improvement in the variation of background can be obtained by the use of external anti-coincidence shields. However, background radioactivity in the sample and scintillator material will still be variable with quench changes.

Other sources of background variations may occur if different sources of chemicals are used in preparation of the scintillator solutions. If solvents made from sources of presently living matter are used as opposed to "ancient" sources (petroleum) the contribution of ^{14}C to the background will be very different. Modern living matter contains about 26 disintegrations per minute (dpm) from ^{14}C per gram of carbon. Thus, in 25 ml of toluene from modern sources there are 20 grams of carbon which will have 520 dpm of ^{14}C. The background from natural tritium also varies depending upon the source of the material. The background levels of tritium in water sources as listed by Tamers [10] are given in Table 6.

Table 6

Tritium Concentrations in Different Water Sources as Taken from Tamers [10]

Water source	Tritium concentration in tritium units[a], T.U.	
	1951[b]	1963
Rain	6	100-1,000
Rivers and shallow lakes	1-6	10-1,000
Underground waters	0-6	0-1,000

[a] Tritium unit is the tritium atoms per 10^{18} hydrogen-1 atoms in the sample.

[b] First thermonuclear weapons test began in 1952.

Conclusions

The use of liquid scintillation counting systems in conjunction with the newly developed combustion techniques and emulsifier systems provides the most efficient method for the measurement of tritium in a wide variety of samples.

ACKNOWLEDGMENT

Part of this work was performed under the auspices of the U.S. Atomic Energy Commission.

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PRACTICAL LIMITS OF TRITIUM DETERMINATION IN A SCINTILLATION COUNTER

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Advances in liquid scintillation technology in the last 10 years have vastly increased the sensitivity and ease with which low energy radionuclides may be assayed. Improvements in multiplier phototube quantum efficiency have quadrupled tritium detection efficiency. Quartz phototube faceplates and graded shielding have reduced background by a factor of 3. Bialkali photocathodes have low background at ambient as well as reduced temperatures providing more flexibility in the choice of reagents.

Development of the sample ratio and external standard ratio provided a convenient means of automating quench correction. New solubilizers, surfactants and scintillators allow a wider variety of sample materials to be counted more efficiently.

In order to effectively utilize the higher sensitivity, special techniques and precautions are necessary. Chemiluminescence, phosphorescence, electroluminescence and static electricity are particularly troublesome when counting tritium. Effects of temperature and humidity variations on measurement stability and variations in background due to sample vials, caps, volume, scintillator and environmental radioactivity are discussed.

Low level counting requires not only low but reproducible background. Good sensitivity requires not only high detection efficiency but determinable efficiency.

Apparatus, Vials, Standards and Reagents

All temperature-controlled measurements were performed in Nuclear Chicago Mark II Liquid Scintillation systems at 10°C. Ambient measurements were taken with Nuclear Chicago ISOCAP/300 liquid scintillation systems including high performance shielding (standard on Mark II). Both systems employ twin phototubes, type EMI 9750QB, with 35 nanoseconds coincidence resolving time. Signals from each phototube are summed before all pulse height analysis.

Unless specified otherwise, "tritium window" is defined as having a 29:1 dynamic range of pulse height, with upper discriminator set at the upper edge of an unquenched tritium spectrum. The lower discriminator is thus set at 18 keV/29 = 0.26 keV, half the average energy to produce the minimum two-photon coincidence. Over 99% of the maximum integral tritium events are included in this window at any degree of quench.

All vials, standards and reagents (except Triton X-100 from Rohm and Haas) were supplied by Amersham/Searle Corporation. Samples were prepared in type 3326 glass, type 3334 nylon, and type 3327 polyethylene vials and capped with type 3367 aluminum caps with polyethylene liner. Unquenched sealed tritium standard Nos. 181340 and 180050 having a calibration accuracy of 3% were employed for efficiency determination and quench standardization. Prepared samples had a calibration accuracy of ±3%.

Background Components

Analysis of the constituents of background are summarized in Table 1. An ambient counter was used to demonstrate the stable low background obtainable without cooling. The empty chamber rate and tritium efficiencies are typical of cooled or ambient systems. Temperature-controlled systems may differ unit to unit from 2.2 to 4.7 cpm and 60 to 69% efficiency, respectively. Ambient systems may have 1 to 2 cpm higher background depending upon specific phototubes and ambient temperature.

Scintillations in the phototube faceplate and accidental coincidences are the major sources of instrumental background in the tritium window. To minimize scintillations produced by both internal and external radiation, a phototube faceplate should have

Table 1

Components of Background

Component	Contribution
Empty chamber	3 cpm
TOLUENE (Solvent)	0.12 cpm/ml
POP-POPOP (Solute)	1 cpm/15 ml
Glass vial	16 cpm
Nylon vial	6 cpm
Polyethylene vial	5 cpm
Standard ampoule	13 cpm

In an ISOCAP/300 preset for ^{3}H (29:1) at 25°C
^{3}H efficiency = 63% (unquenched)

low radioactive contamination, poor scintillation efficiency and low mass. Systems with quartz phototube faceplates satisfy this criteria [1]. In addition, optical coupling to the phototubes through air rather than by light pipes avoids another source of background from Cerenkov radiation [1].

Tritium efficiency using an unquenched standard is given for reference in Table 1. It is the most universal measure of detection efficiency in a liquid scintillation system and should be specified in background comparisons, rather than instrument types. The performance of instruments of the same type may differ considerably particularly if the model is long-lived. Background contributions in Table 1 were obtained from measurements of an empty chamber, empty vials, vials filled with solvent and finally, vials filled with solvent and scintillator.

Vial contributions were determined by subtracting the empty chamber count rate from the average of 10 empty vials. The standard ampoule contribution was similarly determined from a sealed and capped but empty ampoule used for preparing standards. Plastic vials contribute 10 cpm less than glass vials. Relative to weight, however, they are about equal at 1.3 cpm/g. The standard glass ampoule measured 3 cpm less than a glass vial because it has less mass. However, mass alone is insufficient for determining background count rate. Radioactive contamination [1] and scintillation efficiency of the material are additional parameters which influence background.

The background contributed by toluene was an order of magnitude less per unit weight than that of the vials. Nylon and glass vials containing 5, 10 and 15 ml of scintillation-grade toluene were measured for 200 minutes. The average increase in background was 0.12 cpm/ml. There was no detectable difference whether toluene was added to glass or nylon vials. The count rate appeared linearly related to solvent volume. Addition of 6 g/l 2,5-diphenyloxazole (PPO) and 0.03 g/l 1,4-bis [2-(5-phenyloxazolyl)] benzene (POPOP) produced no detectable increase in tritium background at 15 ml. However, the count rate above the tritium window increased by 1.3 cpm/ml of scintillator solution.

Scintillation-grade dioxane in polyethylene vials measured 0.26 cpm/ml in the unquenched tritium window, twice as high as toluene. Chemiluminescence, a potential source of background in dioxane samples, was eliminated as a possible source of the increase in this case. Both single phototube background rates were normal. The higher background with dioxane is attributed to quenching. Tritium efficiency determined from an external standard ratio (ESR) measured 29% for dioxane compared to 58% for toluene, see Figure 4. The increase in background with quench is caused by scintillations from environmental radiation quenching below the upper discriminator of the tritium window.

Background Stability

Phototube instability, environmental radioactivity, phosphorescence, chemiluminescence and static electricity may contribute to background instability in the tritium window.

Three causes of phototube instability are:

1) Variation of thermionic emission with temperature.
2) After-pulses and bursting.
3) Electroluminescence and arcing.

Historically, cooling has been employed to reduce and stabilize thermionic emissions from the photocathodes to a level which produces an insignificant random coincidence contribution. Present-day phototubes have bialkali photocathodes with sufficiently low thermionic emission to produce less than 1 cpm random coincidences at 25°C. Ambient variations from 20°C to 30°C usually change background less than 2 cpm depending upon the specific phototubes used. The empty chamber rate declined 0.8 ± 0.8 cpm or 0.5 cpm/°C, when temperature was reduced from +10°C to -4°C. If background stability better than ±0.5 cpm is desired, temperature stabilization at 10°C or less is recommended.

Some phototubes tend to after-pulse [2,3] due to ion or optical feedback. After-pulses may in turn, generate more after-pulses, creating a burst of pulses. Usually, the bursts go undetected in the single tube background rate. However, the probability of accidental coincidences is high when bursting occurs because of the high burst frequency and time correlation. In this manner high energy events (i.e., from external radiation) may subsequently produce counts in the tritium window even though the primary pulse was excluded by the upper discriminator.

After-pulses are rejected by producing a blanking interval (1 to 4 μsec) after each pulse from either or both phototubes. The blanking interval is retriggerable and, thus, may be extended indefinitely in the presence of bursting. The small loss in counting time is corrected by simultaneously interrupting the timer. In this manner, accumulated time if "live" time.

Electroluminescence [4] and arcing [3] on the phototube envelope may contribute to background instability. Glass tends to luminesce when subjected to a potential gradient. The gradient between pin connections on the base of the phototube can be locally increased in the presence of high relative humidity.

As water molecules adhere to the surface, the potential gradient redistributes, producing regions of high and low potential. Sufficient photons may be generated by electroluminescence or arcing to be detected by both phototubes or be randomly coincident with pulses from the other phototube. Pulse height analysis shows an increasing abundance of single photoelectrons as relative humidity increases. Cooled systems rarely experience this type of electroluminescence because the air is dehumidified upon cooling. The susceptibility of ambient systems to electroluminescence depends upon the type of phototubes, operating voltage, relative humidity and temperature excursions. Operation of the phototubes at low voltages (less than 1500 V) minimizes the electroluminescent effect.

Background stability of an ambient system was determined by measuring the empty chamber count rate in the tritium window for 5000 minutes. It passed Pearson's chi-square test [5] at an expected one sigma deviation of 0.05 cpm.

Increases in environmental radioactivity have less effect in the tritium window than above it [1]. Two inches of virgin lead and steel effectively shield the detector from environmental radioactivity in the tritium window. As shown from the previous background measurements, unquenched background increases 0.12 cpm/ml in the tritium window compared to 1.3 cpm/ml above the tritium window. Quenched samples may be twice as high, see Figure 4. Thus, radiation which has sufficient energy to penetrate the shielding usually produces large pulses which are rejected by the upper discriminator [1]. Nonetheless, regions of high environmental activity like Denver may experience an increase in the background by as much as 0.7 cpm/ml. For example, when an unshielded 4 µCi (Ra 226) source was placed 26 inches from the detector, perpendicular with the phototube axis, the background increased an additional 0.16 cpm/ml. Performance is usually improved by using a lower setting for the upper discriminator in high-radiation environments.

The sample or its container may phosphoresce [16] after illumination under ultraviolet light, emitting single photons at a sufficiently high rate to produce substantial accidental coincidences. An unpredictable increase in tritium background can result from long decaying phosphorescence. White urea caps, standard on most vials, have been found to produce intense phosphorescence which decays in less than a minute. Glass vials have low intensity phosphorescence which dissipates in a few seconds. The coincident rate from polyethylene vial phosphorescence becomes negligible within a few minutes, but nylon vials, particularly freshly manufactured ones, may require 30 minutes dark adaptation. Direct sunlight and fluorescent lighting should be avoided in the counting room where, if the lid to the counter is opened, they may excite a sample prior to counting.

Some chemical reactions used in liquid scintillation counting, particularly those involving strong bases to digest biological material, produce chemiluminescence [7,9]. Reactions may continue for days or weeks unless terminated by neutralization or thermally accelerated to completion before counting. Chemiluminescence is monophontonic and, thus, like phosphorescence, produces accidental coincidences. High sample activity is no guarantee of freedom from excessive background. Chemiluminescent samples may produce thousands of accidental coincident counts.

The background produced by chemiluminescence can possibly be reduced to a negligible level by cooling. If cooling does not reduce the rate to a negligible level, it will act to prolong the decay, possibly increasing the total number of background counts accumulated during a long counting run. Therefore cooling may or may not lower the background from chemiluminescent samples.

Chemiluminescence and phosphorescence may be detected by using a separate low energy channel to look for abnormal spectral shape. Either the double ratio technique proposed by Bush [8], or the double dpm technique proposed by Bransome [9], will detect the abnormally high rate from chemiluminescence just above the coincidence threshold.

Recently, Nuclear Chicago announced a "photon monitor" to guard against erroneous data caused by phosphorescence or chemiluminescence. In addition to the normal coincidence detector, the output from one of the phototubes is delayed and used to determine accidental coincidences. The measured accidental count rate is averaged and compared with the gross coincidence rate. If the measured accidentals are a significant fraction of the gross coincidences, the system pauses to allow the accidentals to decay to an acceptable level. If the fractional accidental rate continues above the minimum acceptable level for a considerable time, the sample is rejected.

Plastic vials may tend to become charged with static electricity and accumulate dirt. All vials should be cleaned thoroughly with a damp wipe before placement in the counter to remove dirt and contaminants without electrostatically charging the vials. Sliding or rubbing of samples on insulated surfaces should be avoided because vials tend to become electrostatically charged. Being good insulators, they discharge slowly, producing spurious scintillations.

Sample Vial Background Reproducibility

Ten empty sample vials each of glass, nylon and polyethylene were counted for 100 minutes to determine the background reproducibility from vial to vial. The one sigma deviation measured on the glass vials was ±1.0 cpm exceeding an expected statistical deviation of ±0.5 cpm. Although lot to lot and vendor to vendor variations are expected to be greater, this is typical reproducibility under lot control. With nylon and polyethylene, the vial to vial variations were undetectable in 100 minutes at a statistical precision of ±0.4 cpm. The polyethylene vials finally showed a nonstatistical deviation of ±0.3 cpm after each was counted for 400 minutes. Thus, differences in background from vial to vial set the lower limit of detection at ±1.0 cpm from glass and ±0.3 cpm for plastic vials. Therefore, replicate sample counting is preferred over replicate counting of one sample when sufficient material is available.

The background contributed by the sample vials is little affected by changes in temperature, solution volume or chemical quenchers. However, color-quenched solutions will quench scintillations from the glass vial which pass through the color quencher, usually reducing the background.

Detection Efficiency

Multiplier phototubes have by far the greatest influence on detection efficiency. Quantum efficiency has more than doubled in the last 10 years (quadrupling tritium efficiency) and may well double again. Bialkali photocathodes have peak spectral response at a shorter wave length (3800 Å), more nearly matching primary solutes than the former CsSb cathodes with S-11 response. Thus, optimum collection efficiency may be obtained without a wavelength shifter, providing the sample itself does not quench the primary solute, absorb its emissions, or the concentration of solute required acts as a quencher [10]. Quartz phototube faceplates and aluminum reflectors provide 80% transmission of the spectral emission down to 2800 Å, whereas systems employing pyrex faceplates or acrylic light pipes are limited to about 3200 Å at the same transmission efficiency. However, improvements in optical collection efficiency have been minor compared to phototube quantum efficiency.

Samples with low efficiency are the most affected by changes in system efficiency. Gains and losses occur at threshold. The probability of detecting a large ionizing event which produces many photons is little affected by the loss of a few photons. On the other hand, small scintillations as from quenched tritium depend upon the few photons. A 5% increase in efficiency from 50 to 55% reduces the counting time only 9%, usually not a significant improvement for unattended operation. However, a 5% increase in a quenched sample from 10 to 15% yields a 33% reduction in counting time. At a given precision, improvements in detection efficiency reduce the time to count quenched samples more than unquenched samples.

An increase in scintillator or phototube quantum efficiency produces both an increase in detection efficiency and pulse height, but an increase in gain beyond the second or third dynode increases pulse height but not quantum efficiency. The efficiency of a quenched tritium sample cannot be restored by increasing the gain. An increase in gain may, however, produce a decrease in the counting efficiency in a window due to pulses rejected by the upper discriminator.

The effect of temperature on detection efficiency was measured by allowing a Mark II at minus 4°C to slowly equilibrate at 27°C. Efficiency of an unquenched tritium standard declined continuously from 65.5% to 64.5%, yielding an average temperature coefficient of minus 0.03%/°C. To separate efficiency from gain, the detection efficiency was measured integral at minimum attenuation and discrimination.

Vial and sample geometry also affect efficiency. Diffuse samples with optical transmission equal to a transparent sample count more efficiently than transparent samples [11], i.e., condensation on a glass vial increases tritium efficiency about 2%. Accumulation of dirt in the counting chamber or on the vials has much more influence on tritium than ^{14}C efficiency. Whereas ^{14}C efficiency may decline only one or two percent, tritium efficiency can decline as much as ten percent.

Efficiency variations with sample volume are shown in Figure 2. Although maximum efficiency occurs from light emanating from the center of the vial, the average efficiency in the volumetric integral peaks above 10 ml. Optical diffusion, absorption and color quenching are other examples of geometry-dependent variables which influence efficiency. Unlike chemical quenching, which produces an energy loss independent of location in the sample, geometry-dependent variables produce not only different spectra, but at the same time can produce wide variances in spectral shape [12]. Spectral resolution of luminous intensity is degraded at low intensity by quantum and multiplier statistics. At the lowest limit of detection, when only one photoelectron is produced at each photocathode, the pulse height distribution is independent of quenching mechanism. Only the phototube parameters control the spectral distribution.

Figure 1

Tritium Sample Channels Ratio (SCR)

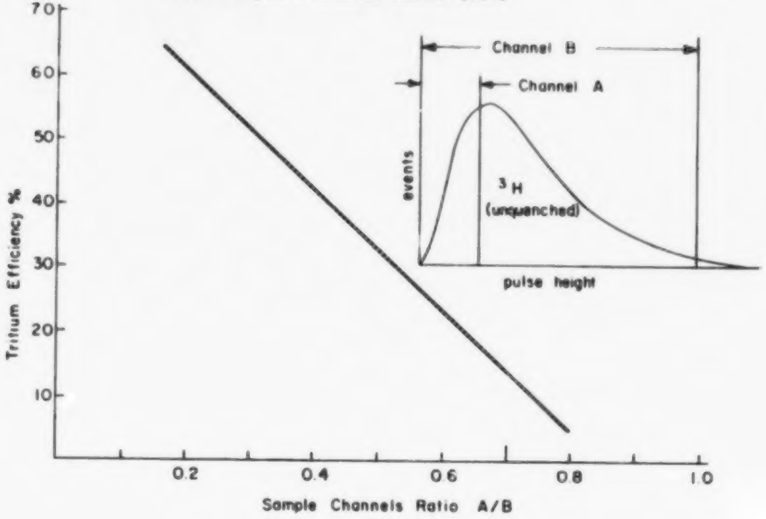


Figure 2

Volume Dependence of Efficiency 3328 Glass Vials in
a MARK II, 29:1

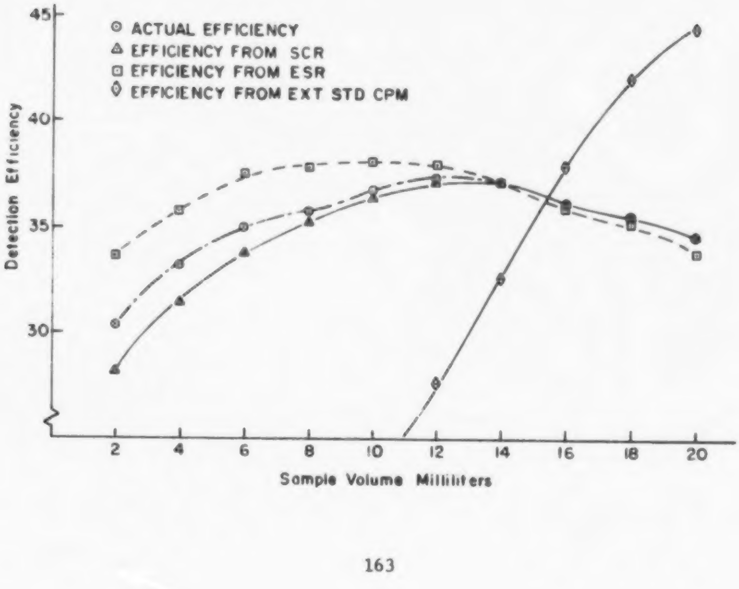


Figure 3

Reproducibility of Measured Tritium Activity from
15 Cycles in a MARK II

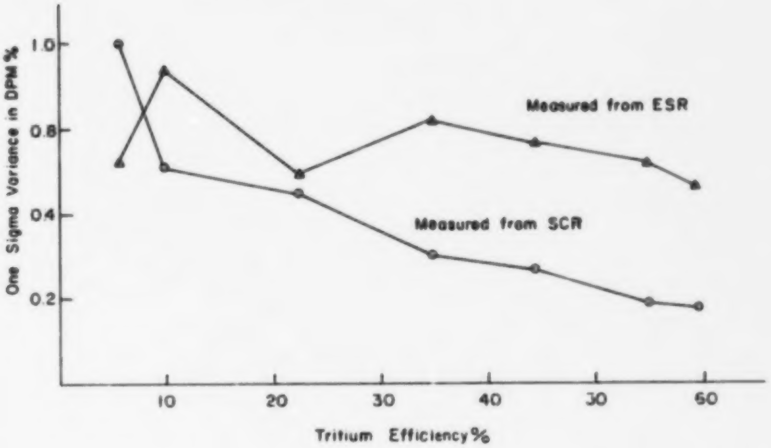
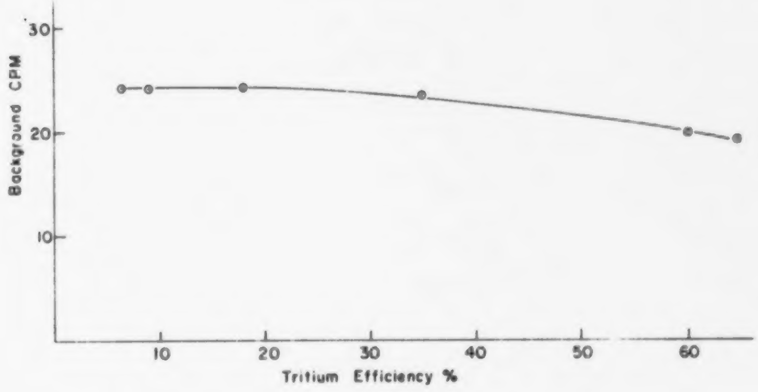


Figure 4

Background vs. Quench 3323 Glass Vials in a MARK II, 29:1



Efficiency Determination

Sample preparation and efficiency determination present the most arduous problems to liquid scintillation methodology. The homogeneity of sample and scintillator, which imparts high detection efficiency and ease of efficiency determination for low energy beta emitters, causes the problem. Samples which are not completely solubilized may result in erroneous efficiency determination. Heterogeneous samples should be avoided, particularly for tritium. Variations in particle size and self-absorption may change detection efficiency in an indeterminable manner [8].

Two related effects of quenching are a reduction in both pulse height and detection efficiency. Baille [13] first described the sample channels ratio method of utilizing the former to determine the latter. Investigation by Bush [14] using different solvents, fluors and chemical and color quenchers determined that for each isotope, one sample ratio quench curve was adequate to determine efficiency, provided the samples were not color quenched. A typical quench curve is shown in Figure 1.

Neary et al. [12] demonstrated that quench is dependent upon path length through a color quencher, whereas chemical quenching is local to the primary event. The pulse height spectrum of color-quenched samples results from a continuum of path length dependent quench, which accounts for its broader, flatter spectrum, thus, the efficiency of colored samples is overestimated using a chemical quench standard curve.

Errors in the activity determined from a chemical quench curve may be as high as 20% when samples are heavily color quenched, particularly if there is no chemical quench. In the limit color-quenched samples yield spectra characteristic of self-absorption which are indeterminable. Earlier phototubes were unable to resolve these spectral differences in tritium samples because of low quantum efficiency. Sample volume, absorption and re-emission in the solute, and light scattering are other examples of parameters which, if changed, may alter the spectral shape.

If colored samples are to be processed routinely, a separate quench curve should be constructed, preferably with actual samples. Activity of the sample standards may be determined by dilution, extrapolation or by internal standardization. Mildly colored samples may deviate little from the standard chemical quench curve. Curves of heavily colored samples may not be valid for other sample-solvent systems unless the ratio of color to chemical quench is the same.

At low count rates the contribution of the background spectrum may be significant. Subtracting it from the analysis and SCR channels (if used) before calculating the ratio is usually sufficient for tritium because changes with quench are gradual, as shown in Figure 4.

Excess phosphorescence and chemiluminescence produce unwanted, accidentally coincident counts just above the coicidence threshold. Not only is the apparent count rate increased, but the overall spectral distribution is altered so as to appear more quenched. Activity calculated from the sample channels ratio is thus doubly overestimated by counts added to the cpm and apparent shift in ratio.

Frequently samples of different volume are counted. Errors in efficiency, determined from three methods of quench correction, are shown in Figure 2:

1. Sample channels ratio, SCR (Figure 1)
2. External standard ratio, ESR
3. External standard cpm, ES

Both the SCR and ESR methods track the actual efficiency within ±1% from 12 to 20 ml, whereas efficiency determined from the ES cpm method changes in the opposite direction of the actual change with volume, generating an error of 2.5% ml. The number of Compton electrons produced by the external standard depends upon electron density as well as volume of the sample. The ratio method effectively normalizes the count rate, making it independent of external standard count rate.

The data in Figure 2 were obtained from 10 samples prepared in glass vials. A solution containing 73% toluene, 26.5% ethanol, 0.5% tritiated water, 5 g/l PPO and 0.06 g/l POPOP was added to all samples. Activity of the solution was 9,781 dpm/g. Capped vials were weighed before and after pipetting in 2 ml increments. Actual activity of each sample was determined by weight rather than volume to eliminate pipetting errors. Samples were capped with type 3367 aluminum caps and polyethylene liners. In addition, triplicate samples were prepared in the same manner in 5, 10 and 15 ml polyethylene and nylon vials.

In the nylon vials, the count rate slowly declined due to permeation of the tritiated water through the nylon. After four days, the cpm/g was less than two thirds of the original value, though the SCRs and ESRs remained unchanged. Half of the vials had swelled and become stuck in the sample conveyor. Nylon vials have not generally been recommended for aqueous solution. Although the aqueous sample diffused through the vial walls there was no change in quench indicated by either the SCR or ESR.

The count rate in the polyethylene vials declined about 5% as the average weight declined 1.2 g (13%) in 18 days. The SCR remained constant. Unexpectedly, the efficiency determined from the ESR slowly declined. After six days all samples had stabilized at over 6% loss in absolute efficiency determined from the external standard. The change at 5 ml was larger but slower than at 15 ml. The 15 ml samples stabilized in less than 2 days. No deformation of the vials was observed after 20 days. The shift in only the efficiency determined from the ESR was unexplained by leakage.

Four unlabeled 5 ml samples were prepared in polyethylene vials to examine the change in ESR. Each contained 5 ml of toluene, with 6 g/l PPO and 0.03 g/l POPOP, respectively. The Compton spectrum from Ba-133 and Ra-226 external standard sources were examined with a multichannel analyzer. All three of the blanks containing toluene increased in count rate with time at low energy whereas the dioxane blank remained unchanged. Toluene blanks containing scintillator increased about 70,000 cpm whereas pure toluene increased only 33,000 cpm at one third the pulse height. Contents of the three vials were removed and counted separately in glass vials with the external standard. No abnormal counts nor spectral shift were observed. The three empty polyethylene vials retained the increase in count rate after twice flushing with ethanol. The spectrum was at one third the pulse height of the unquenched solution for both Ba-133 and Ra-226 external standard sources. Thus, polyethylene probably behaves like a fluor in the presence of toluene. As toluene diffuses into the vials, scintillation from Compton electrons in the vial wall increases in intensity and pulse height. Compton electrons from the vial appear quenched and thus alter the external standard ratio toward more quench. At low sample volumes, the effect is more pronounced because fewer Compton electrons are produced in the sample solution. The shift in the ESR from polyethylene vials containing toluene is caused by an increase with time in the scintillation efficiency of the vial walls as solvent diffuses into the wall. The effect is independent of the external standard source. The increase in count rate is mostly in the low energy channel used for the ESR. The added spectrum from the vial is proportional to the energy of the external standard, independent of the external standard source.

One should thus avoid external standardization of samples containing toluene in polyethylene vials. If the toluene-polyethylene external standard combination cannot be avoided, samples should be filled to 20 ml and stabilized for two days before counting. Quench correction by external standard count rate could be achieved by selecting an external standard window above the vial spectrum. In addition to the inherent sample volume (Figure 2) and electron density sensitivity of the cpm method, the useful range of quench would be limited to a three to one change in pulse height. This would severely limit present instruments which are designed to accommodate at least a 25:1 reduction of pulse heights.

The polyethylene vials were permeable to toluene but not to methanol or water. Tritiated toluene count rates in polyethylene declined about 1/2% a day. Weight loss the first day was small until the toluene had diffused through the wall.

Reproducibility of the activity determined from the sample channels ratio, SCR, and external standard ratio, ESR, methods of quench correction are shown in Figure 3. Data for cpm were collected at a statistical precision of ±0.1%. The resultant activity uncertainties are due to the statistical precision of the method and its ability to track small differences in efficiency caused by repositioning of the sample. SCR data were accumulated for the same time it took to accumulate the cpm data. Measured variances in the dpm obtained from the SCR are slightly greater than the expected statistical variance, while that obtained from the ESR is about 0.3% greater than expected. Note that a relative uncertainty of 1% at 10% detection efficiency is equal to an absolute uncertainty of 0.1% efficiency.

Performance of Triton X-100

A 20 ml sample containing 6 ml tritiated water was prepared in a polyethylene vial using the technique described by Williams [14]. No attempt was made to optimize solvent, solute or water concentrations or to add a secondary solute or to optimize windows. The 14 ml scintillator cocktail contained 35% Triton X-100, 65% toluene and 10.5 g/l butyl PBD. An identical sample containing tritiated toluene and distilled water was used to check sample efficiency. A third unlabeled aqueous sample was used to determined background.

Samples were counted at 10°C in two windows. The first was set at minimum attentuation in a 12:1 window corresponding to attenuator and discriminator settings of A000 and 3 to 35. The "H-3"/pushbutton (dual label tritium) was selected in the second channel. Results shown in Table 2 demonstrate higher figures of merit for aqueous samples using Triton X-100 than previously published [6,15] due to the higher sensitivity of recent liquid scintillation counters.

Table 2

Tritiated Water - TRITON Figure of Merit*

Window	A000 3-35	A200 3.5-50
Efficiency	28.9%	29.5%
Background**	8.45±0.1	10.9±0.1
E^2/B	98.8	79.8
$EVB^{-1/2}$(V in ml)	59.6	54.1
EV (V in %)	867.0	885.0
M.D.A.***(30min)pCi/ml	0.28	0.31
T.U./cpm	64.0	64.0

*Sample composition: 14 ml scintillator and 6 ml water in a polyethylene vial.
Scintillator: 35% TRITON X-100, 65% Toluene, 10.5 g/l But-PBD
Instrument: NUCLEAR-CHICAGO MARK II at 10°C

**With contemporary water

***M.D.A. = 90(B/T)1/2/EV pCi/ml [1].

Pulse height spectra of the water-Triton samples showed the background having a longer, flatter tail than the tritium spectrum. One could thus adjust the upper discriminator for maximum performance where background is crucial. About one fourth of the background is instrumental, and one third of it from the vial.

Activity of the aqueous samples measured by the SCR method was under estimated about 8% using quench curves from sealed standards. Optical diffusivity of the sample is the most probably cause. Diffused samples require separate quench curves for accurate efficiency determination.

Although the actual efficiency declined exponentially 3% in 64 hours, the calculated dpm increased only 3% in the first 30 hours. For the next 34 hours the dpm calculated from the SCR was stable. The one sigma variance measured ±0.26%. Forty-eight hours later the sample measured within 0.5%. After one hour of stabilization at 10°C the tritiated water sample was within 0.7% of the efficiency of the tritiated toluene sample.

Conclusions

A modern liquid scintillation counter may be expected to reproduce a tritium assay within ±2% at one sigma over a wide range of quench and sample volume, heterogeneous and color quenched samples excluded. Reproducibility of ±0.5% is possible at efficiencies greater than 20% using the SCR method of quench correction. However, total counting time of low activity samples may be longer than with the external standard.

Sample vials are the major source of background uncertainty: glass ±1.0 cpm, polyethylene and nylon ±0.3 cpm. Prior determination of vial background or statistical averaging can reduce the uncertainty in background. Excess background due to phosphorescence and chemiluminescence is detectable by at least three methods: double ratio, double dpm, and photon monitor. The latter method automatically takes corrective action by delaying or rejecting an errant sample. Temperature stabilization is recommended when background stability better than ±0.5 cpm is required. Higher figure of merit, E^2/B, is obtained by setting the upper discriminator to a lower value in the presence of high environmental radioactivity.

Tritium sensitivity in a liquid scintillation counter now equals proportional counters. The ease and reproducibility with which an experimenter may assay tritium in a liquid system makes it the method of choice. Tritium samples containing 6 ml of water can be counted at 30% efficiency with 64 cpm T.U. Higher sensitivity may be attainable by optimization of fluors and windows.

ACKNOWLEDGMENT

The author gratefully acknowledges the guidance of Mr. D. L. Hansen in designing and preparing experiments, and Mr. H. Engberg for assistance in the measurements.

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CONTINUOUS MONITORING OF AQUEOUS TRITIUM ACTIVITY

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Development of flow cells, applications, and the need for higher efficiency, particularly for weak β-emitters, has been reviewed by Schram [1], who classified cells into three types: external counting cells where the cell is or is in contact with a plastic scintillator, cells packed with fluor particles or crystals, and homogeneous counting systems where the sample is continually mixed with a liquid scintillator. For weak β-emitters such as tritium, the homogeneous counting system gives much higher efficiency.

A flow cell of the homogeneous counting type was developed and evaluated for tritium monitoring. Parameters which affect performance, such as volume, flow rate, mixing of sample with scintillation solution, contamination and light leaks were investigated; where problems existed, solutions were found. One objective of the present work was to develop a system for monitoring secondary reactor coolant. Performance of the flow cell for this application was tested.

Description of Flow Cell System

Figure 1 is a diagram of the system used in our studies. Scintillator solution flows from the constant-head reservoir through a glass capillary restrictor (which determines the flow rate) to the mixing chamber. Radioactive sample is introduced into the mixing chamber via sample inlet. The mixed solution flows through a light trap, a probe assembly, and finally a flow cell in a β-Mate II counter, from which it flows through a second light trap to waste.

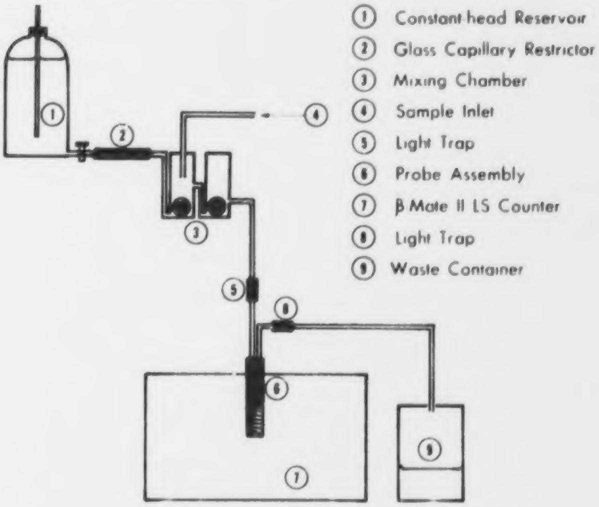


Figure 1

Continuous Flow Cell and Liquid Scintillation System

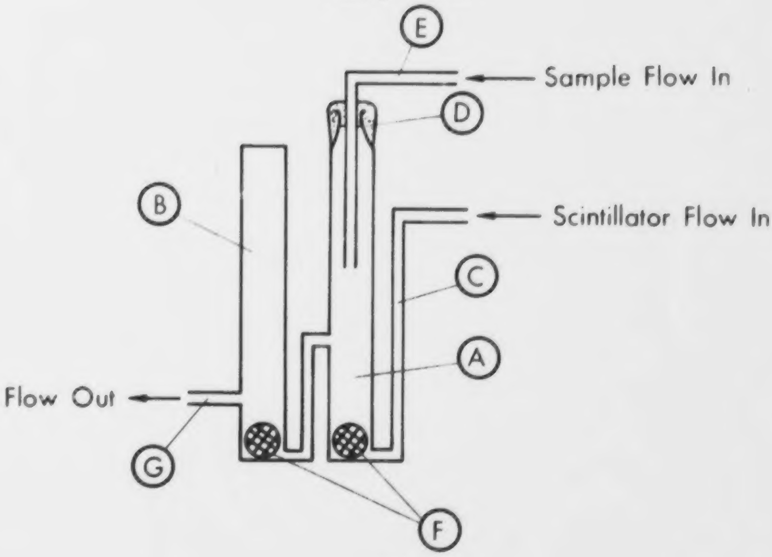


Figure 2

Mixing Chamber

The mixing chamber is shown in Figure 2. Two glass cylinders, A and B, are closely connected. Scintillator solution flows into cylinder A from the tube C. Cylinder A terminates in a "pop-bottle" opening which is closed with a rubber serum-cap D, through which an inlet tube E is inserted for introduction of sample. The use of the serum-cap closure provides for quick exchange of inlet tubing E. In addition, a serum cap can be fitted without inlet tubing, to serve as an injection port for the introduction of measured volumes of batch samples by means of a syringe. This allows for quick assay of batch samples on a LS counter without the necessity of removing the flow cell assembly. Each cylinder contains a spherical encapsulated magnet F for stirring by a standard laboratory magnetic stirrer. The combined sample-scintillator solution exits from B via the side arm tube G.

During operation, the mixing chamber is filled with scintillator solution to the levels indicated by the outlet tubes. Sample is ejected dropwise from tube E, the drops falling into and mixing with the stirred liquid in A. Flow of the mixture through the second stirred volume in cylinder B assures complete homogeneity.

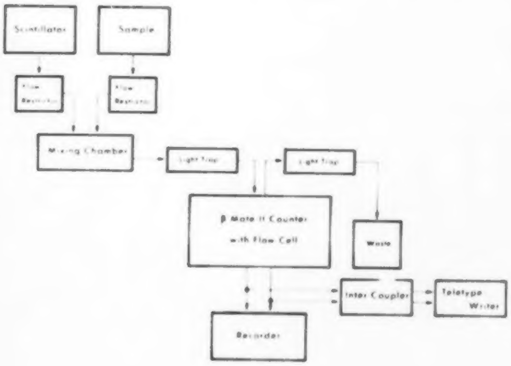
In the course of our studies we encountered the problem of light piping. Light may be piped into the flow cell by traveling through the liquid media via reflection from the inside surface of the connection tubing. This light piping produced high background.

A simple and effective light trap for eliminating this problem was developed. The light trap consisted of a coil of glass tubing with an irregular inner surface. The light trap and the connecting tubing between the trap and the flow cell were protected from direct light entry by painting with black paint. In order to eliminate light piping into the cell from both ends, two light traps were used in the flow cell system.

All experiments were conducted with a Beckman Continuous Flow Cell β-Mate II System. Beckman Ready-solv^{TM} VI and 1,4-Dioxane with Beckman Fluoralloy Dry Mixes were used as scintillation solvents for aqueous samples.

Figure 3

Block Diagram of the Experimental Arrangement



The ratemeter output of the counter was displayed on a Beckman 10" recorder and also was converted to digital form via an intercoupler and teletypewriter. Data were processed on a Wang 720 A computer.

A schematic block diagram of the overall instrument arrangement is shown in Figure 3. The scintillator and mixed solution were gravity flow. Since continuous monitoring experiments were conducted under simulated conditions, reactor secondary coolant and other tritium-labeled water samples were introduced into the mixing chamber by means of a peristaltic pump.

Results and Discussion

Injected samples were used to study the continuous flow cell system. Results revealed the fundamental characteristics and capabilities of this system.

Analog recording of one drop response is shown in Figure 4. The drop was introduced into the mixing chamber when background level remained constant. The activity peak rose sharply and went down slightly slower. Residence time was less than two minutes with this particular flow cell, 4.3 ml volume, and scintillator flow rate, 7.0 ml/min. Under these conditions, peak height system counting efficiency for tritium was about 10%.

Figure 5 is an analog record of five successive injections of tritium-labeled water samples of 10, 20, 30, 40, and 50 microliters, respectively. The ratemeter used in these measurements had a logarithmic output. In this case the most accurate method of reading cpm is to read the counter on the instrument. This was the method used to obtain cpm at peak height maximum. An alternative method was used for Figure 6, which is a plot of the counting data for peak 5. Counting data were input directly to the Wang 720 A computer through an intercoupler. This system gives a linear plot of time versus count rate. The two negative values shown in this plot arise from statistical variations in the data near the background count rate.

Count rate vs. sample size of the five successive injections is plotted in Figure 7. The response to change of activity by this system is linear within the statistical counting error. Tests were also made to establish the maximum sample size to be introduced into the system without causing a mixing problem. Results indicated that maximum sample flow rate for continuous monitoring was dependent on configuration and volume of the mixing chamber, scintillator flow rate and the scintillator solution. Under optimum conditions, continuous sample flow rate could be as high as 0.6 ml/min.

Scintillation Flow Rate Vs. Efficiency and Response Time

For all flow cell systems where scintillation liquid is added, dilution occurs. The degree of dilution is a function of scintillator flow rate, sample flow rate (sample size for injected samples) and volume of the mixing chamber. Minimum dilution under ideal conditions is determined by the solubility limit of the sample in the scintillation liquid, or by the amount of sample which can be mixed and maintained in a homogeneous suspension in the scintillation liquid.

In our studies, we have tried several versions of mixing

Figure 4

Response to One Drop Sample
(Drop size 0.04 ml, tritium labeled water sample 8.18 X 10^4 dpm/ml, volume of the flow cell 4.3 ml, scintillator flow rate 7.0 ml/min)

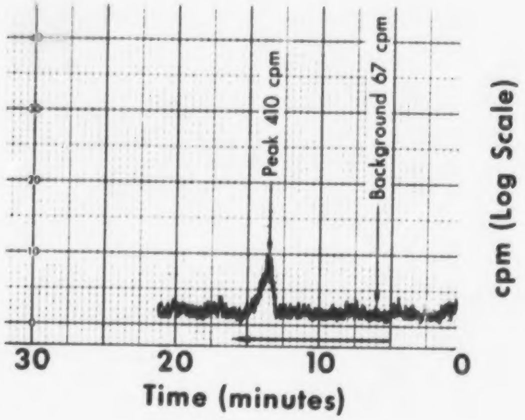


Figure 5

Analog Recording of Ratemeter Output for Five Injected Tritium Water Samples

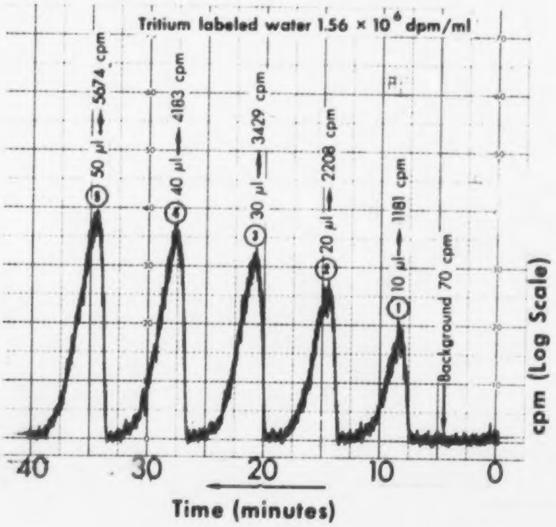


Figure 6

Computer Output Corresponding to Peak 5 in Figure 5

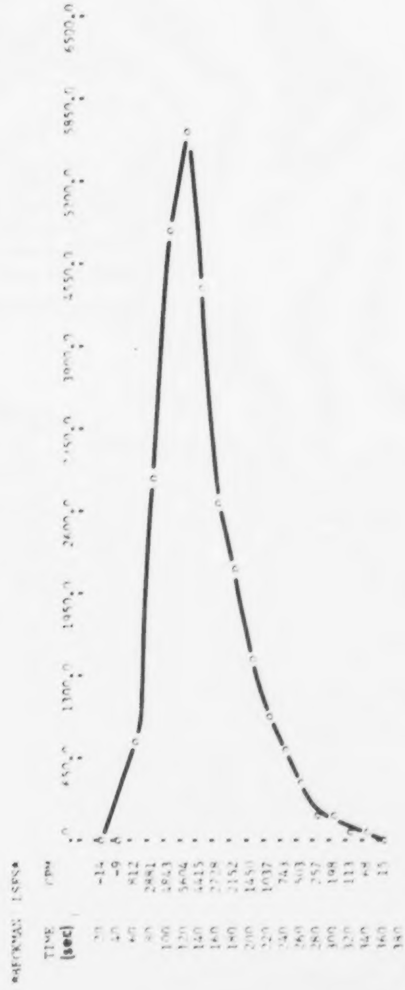
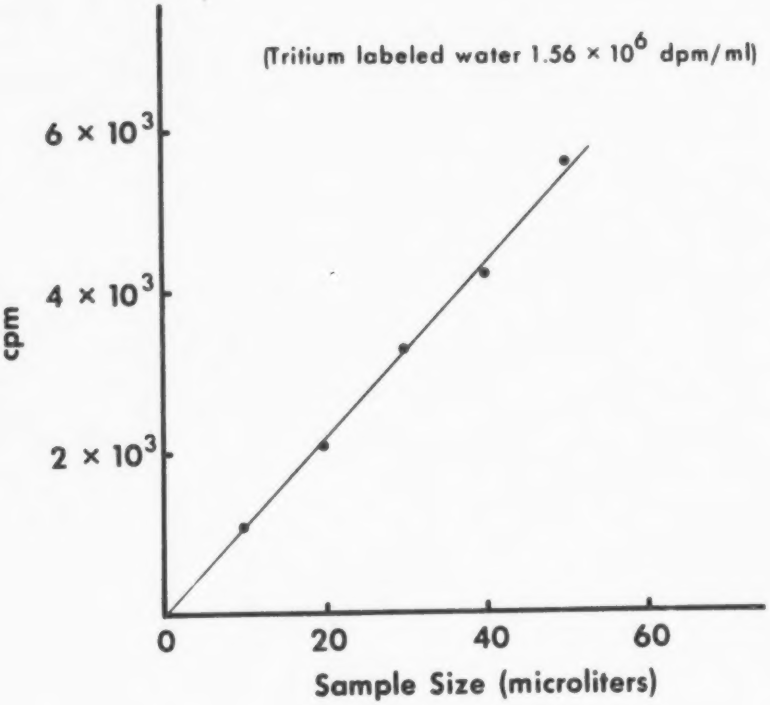


Figure 7

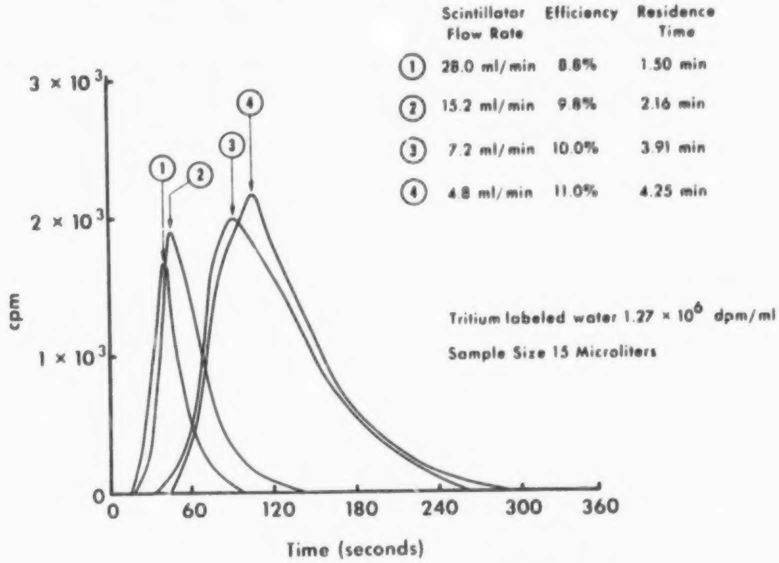
Count Rate vs. Sample Size



chambers. The final version described earlier was the optimum configuration and size for maximum efficiency with a reasonable residence time. With this mixing chamber, four samples of the same activity were measured at different scintillation flow rates. Results are shown in Figure 8. Although peak height counting efficiencies were about the same, response time was inversely proportional to scintillator flow rate. This would be the case for discrete samples with a volume less than required to reach maximum efficiency. For continuous samples the ratio of scintillator flow to sample flow will affect system efficiency. System efficiency is maximum whenever scintillator flow to sample flow is minimum. This leads to the selection of low scintillator flows. However, if it is desirable to minimize response time, then the scintillator flow should be increased. If sample flow is increased accordingly, the only disadvantage is the increased use rate of scintillator.

Figure 8

Scintillator Flow Rate vs. Efficiency and Residence Time



Reproducibility

In order to obtain good reproducibility, the scintillator and sample flow rates must be kept at a constant ratio, mixing must be complete and homogeneous, there must be no light leak, and the counter itself must be reliable.

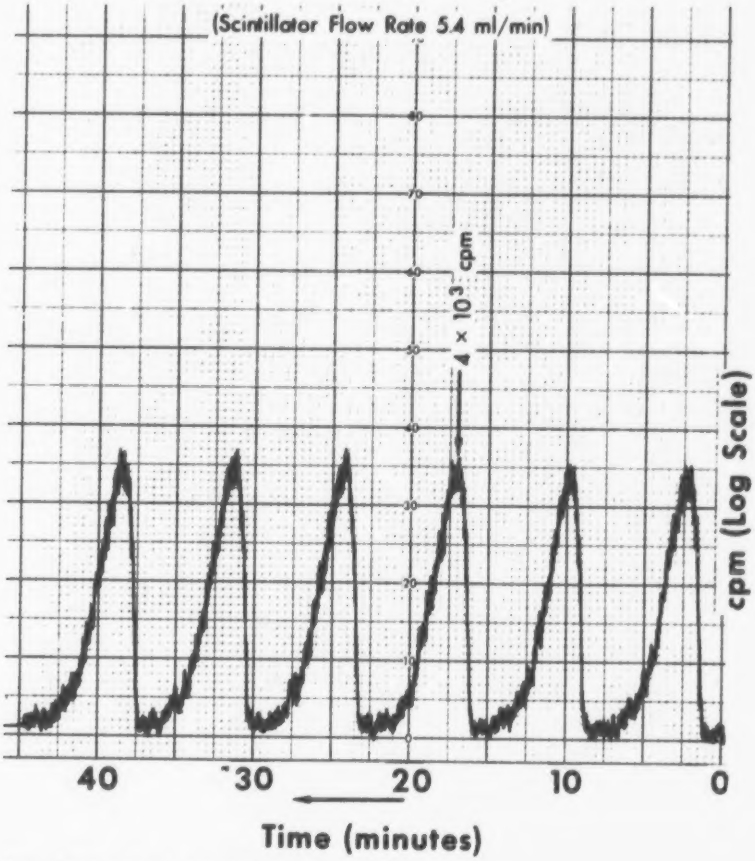
Although several methods of regulating the scintillator flow rate were tested, the simplest and most reliable method was to use a glass capillary tube. Various lengths were used for different flow rates. Sample flow is regulated in the same way.

In addition to rapid and homogeneous mixing, the properties of the scintillator solution must also be considered. For most aqueous samples, Beckman Ready-Solv™ VI was found suitable and is preferred because of higher counting efficiency. However, for low activity samples, such as reactor secondary coolant, maximum sample flow rate is necessary. In this case solubilization is not fast enough for homogeneous mixing. Therefore dioxane-based scintillator solution was used. The disadvantage of dioxane-based scintillator is slightly lower counting efficiency for tritium compared with toluene-based Ready-Solv. However, for this application, increased solubility more than compensates for decreased efficiency.

Figure 9 is an analog record of six successive injections of 40 μl. Dioxane-based scintillator flow rate was 5.4 ml/min. Repeatability was good under these conditions, and results also showed

Figure 9

Analog Record of Six Successive Injections



that there was no contamination or memory problem.

Continuous Flow Monitoring Studies

Efficiency

In static counting, the efficiency (E) of a liquid scintillation counting system is determined by E = cpm/dpm, where cpm = counts per minute, and dpm = disintegrations per minute of the radioactive material in the counting vial.

The cell efficiency of a continuous flow cell is calculated in the same general manner but, if the efficiency is determined with a mixed flowing sample, it is necessary to take the dilution factor and cell volume into account. Dilution factor (D) is defined as follows:

$$D = \frac{\text{Scintillator flow rate + sample flow rate}}{\text{Sample flow rate}}$$

then

$$E\ (\text{cell}) = \frac{\text{cpm}\ (D)}{\text{dpm}\ (V)}$$

where

V = Cell volume

For the cell which was used in this work, cell efficiency was determined with a flowing sample.

A sample activity of 4250 dpm/ml with a scintillator flow rate of 6.0 ml/min (dioxane base), a sample flow rate of 0.5 ml/min, volume (V) of 4.3 ml, resulted in a count rate of 450 cpm.

$$E\ (\text{cell}) = \frac{450\ \dfrac{(6.0 + 0.5)}{0.5}}{(4250)\quad(4.3)} = 32\%$$

The value of 32% is the highest efficiency which could be achieved for ^{3}H in a dioxane-base scintillation liquid. In actual practice it is *system efficiency* or counting capability which is of primary importance.

With a continuous sample of constant activity, system efficiency depends on cell efficiency, dilution factor and cell volume.

$$E\ (s) = \frac{E\ (\text{cell})\ (V)}{D}$$

when

E (s) = system efficiency

In a typical application, such as reactor coolant monitoring where scintillator flow was 4.8 ml/min and sample flow was 0.6 ml/min, system efficiency with 4.3 ml cell should be

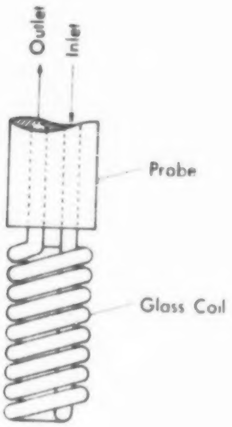
$$E\ (s) = \frac{32\%\ (4.3)}{9} = 15.3\%$$

With these flow rates, cell volume and system efficiency, the detection limit of this system calculated at 95% confidence level is 120 pCi/ml, and at 67% confidence level is 60 pCi/ml.

With a real sample of reactor secondary coolant which had an activity of 2530 dpm/ml, the continuous monitoring record shown in Figure 12 showed an efficiency of 15.4%. Note that, in an application of this type, efficiency of the system is flow rate independent, but would vary with cell volume and dilution factor. If batch samples or varying activity samples are counted with the continuous flow system, system efficiency is variable and calculations are more complicated.

Figure 10

Flow Cell (Effective Volume 4.3 ml)



In this case the sample is contained in a greater volume in the cell than is indicated by the dilution factor obtained by the ratio of total flow to sample flow. Minimum peak height system efficiency is obtained with the smallest sample and highest scintillator flow. This condition was approximated, and a peak height system efficiency for tritium of about 8% was obtained. With larger samples and slower flow rates, this efficiency would tend to increase to about 15%, the continuous sample introduction value.

Efficiencies were determined with cell shown in Figure 10. The effective volume of the flow cell was 4.3 ml. Maximum efficiency was obtained when the glass coil was exposed without any protective covering. Comparative tests were made with the coil encased inside a standard glass vial and measurements were taken when the vial was filled with scintillator solution and when it was empty. Results in both cases showed lower efficiency than the coil without a vial.

Response to Change of Activities in Continuous Monitoring

Two tritium water samples with different activities and a water sample without activity were introduced into the system successively for checking response to change of activities in continuous monitoring. Scintillator flow rate and sample flow rate were kept constant at 4.8 ml/min and 0.6 ml/min, respectively. Background was about 50 cpm. Results are shown in Figure 11. Memory or contamination of the system was not a problem. Maximum response to a change in activity was achieved in three minutes or less. This corresponds to a flow of less than four cell volumes.

Figure 11

Continuous Monitoring Response to Change of Activity

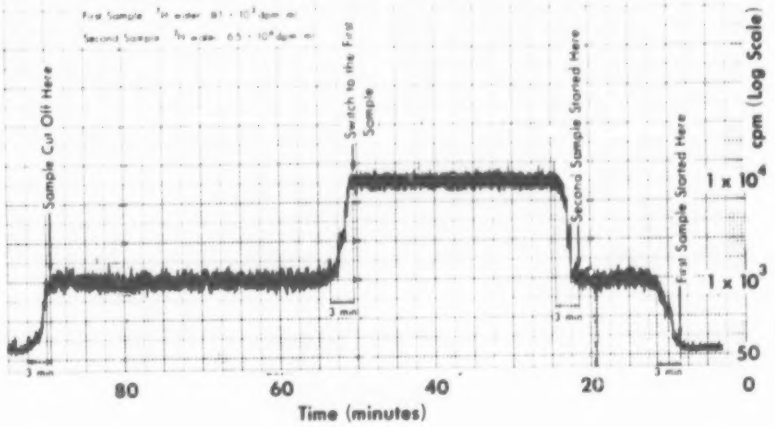


Figure 12

Reactor Secondary Coolant Continuous Monitoring

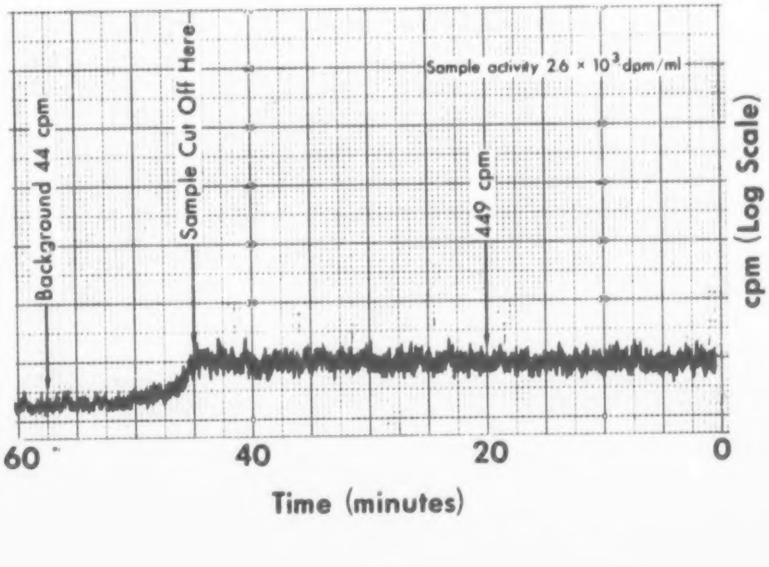
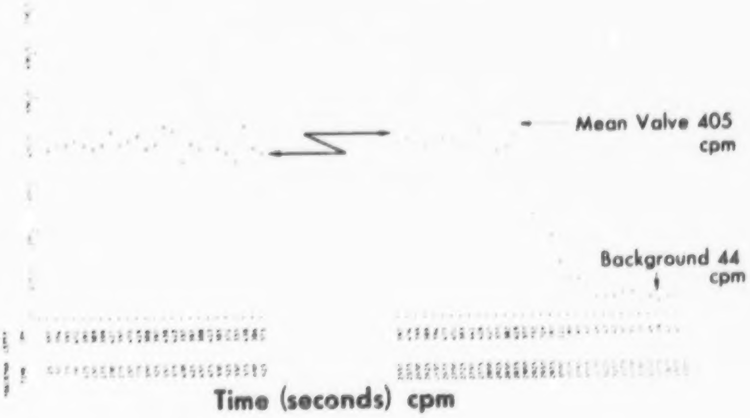


Figure 13

Computer Results of Reactor Secondary Coolant Monitoring



Reactor Secondary Coolant Monitoring

Tritium level of a typical reactor secondary coolant was determined by this system. Sample was introduced into the mixing chamber at 0.6 ml/min by a peristaltic pump, and dioxane-based scintillator flow rate was maintained at 4.8 ml/min. Averaged tritium activity was found to be 1.15 nCi/ml (static liquid scintillation counting showed 1.16 nCi/ml in the same sample). Analog record and computer data reduction results are shown in Figures 12 and 13.

Since the composition of the secondary coolant and many other aqueous stream samples remains relatively constant, the amount of quenching remains relatively constant. Therefore, once the quench condition of the sample is determined, the calibration correction standard can be used throughout the experiment.

Conclusion

A simple, continuous flow cell system has been described and performance evaluated for a weak β-emitter (tritium). Efficiency of the system is high. Contamination (or memory) is low. Less than four cell volumes are required for complete clean-up.

Another common problem, light piping, was also eliminated. With the specially designed mixing chamber, the system is capable of detecting as low as 60 pCi/ml. The serum-cap sample port provides the capability of individual injection. This is convenient and useful if the LS counter is set up for continuous monitoring, but single sample measurements are required occasionally.

The flow cell probe can be inserted into all Beckman Liquid Scintillation System counting wells. No mechanical and electronic modifications are necessary. A standard teletypewriter, paper tape, or a recorder with one hundred millivolt full scale range can be used as the read-out system.

With the growing concern in water pollution, increasing use of radioactive materials in research, and demands in nuclear power plants for safety surveillance, reliable and economical continuous flow monitoring systems for radiation detection are needed. It appears that the described flow cell system meets many of the requirements.

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APPLICATION OF EMULSIONS IN TRITIUM COUNTING WITH REFRIGERATED SYSTEMS

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The use of emulsifiers in the low-level radioassay of tritiated aqueous samples by liquid scintillation counting at ambient temperature was investigated by this Laboratory and a new emulsifier for this purpose, Triton N-101, was introduced [1].

Several investigations have been reported on the use of emulsions for liquid scintillation at low temperatures [2,3]. An optimum mixture reported with toluene-Triton X-100 incorporated 60 percent water with a tritium counting efficiency of 15.5 percent.

The objective of this investigation was to find an optimum emulsifier system that would incorporate 40 percent water and remain clear at temperatures below 10°C. The emulsifiers were evaluated on their ability to produce clear emulsion concentrates with the required amount of water and on the resultant counting efficiency.

Procedure and Results

Individual mixtures in polyethylene liquid scintillation counting vials were tested by refrigerating at 4°C and examined visually for periods up to 48 hours. Those mixtures clearing within 8 hours after the start of the refrigeration and remaining clear up to 48 hours were marked for further investigation.

Based on previous work with emulsions [1,4] this investigation was limited to two primary solvents, toluene and p-xylene. The emulsifiers investigated were Triton X-100 and Triton N-101 because of their superior emulsifying properties with mixtures containing water and xylene or toluene. Triton X-45 was added to the mixtures as a hazepoint depressant in order to reduce the temperature range of the clear emulsion stage. All three emulsifiers are commercially available alkyl phenylpolyethylene glycol ethers, the Triton series being supplied from Rohm and Haas Company. Table 1 shows the results of a number of preliminary tests in which emulsifier-solvent proportions were varied.

It was decided, based on this test, to prepare a scintillation mixture containing fluors using the approximate concentrations of emulsifiers and toluene from mixtures 5 and 7 as shown in Table 1. The counting efficiencies of the two mixtures were found to be identical and mixture 5 was chosen for further examination.

Table 1

Results of Visual Inspection of Emulsions at 4°C
Containing 40% Water

No.	Emulsifier, %	Solvent, %	Physical Appearance	
			8 hours	48 hours
1	Triton N-101, 8.5 Triton X-100, 8.5	toluene, 43.0	Opaque	Opaque
2	Triton N-101, 17.0	toluene, 43.0	Opaque	Opaque
3	Triton N-101, 8.5 Triton X-45, 8.5	p-xylene, 43.0	Opaque	Opaque
4	Triton N-101, 8.5 Triton X-45, 8.5	toluene, 43.0	Opaque	Opaque
5	Triton N-101, 13.0 Triton X-45, 4.0	toluene, 43.0	Clear	Clear
6	Triton N-101, 13.0 Triton X-45, 4.0	p-xylene, 43.00	Opaque	Opaque
7	Triton N-101, 12.0 Triton X-45, 4.0	toluene, 44.0	Clear	Clear

The following mixture was prepared based on proportions from experiment 5: Toluene, 72.0%; Triton N-101, 21.2%; Triton X-45, 6.8%.

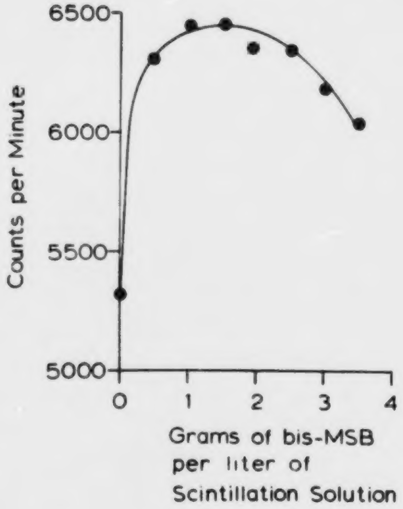
Counting experiments were set up to determine the effect of secondary fluor, p-bis-(o-methylstyryl)-benzene, (bis-MSB), on the counting efficiency to the above mixture when used with 40 percent water. Based on these experiments, 1.5 grams of bis-MSB were used as the optimum amount for 1 liter of toluene mixed with 390 ml of emulsifier blend. The primary fluor was diphenyloxazole.

The final mixture used to incorporate 10 ml of tritiated water for each addition of 15 ml of scintillation solution was the following:

Toluene	1000 ml
Triton N-101	296 ml
Triton X-45	94 ml
PPO	7.9 g
bis-MSB	1.5 g

Liquid scintillation solutions based on the above composition were prepared and counted on a Packard TriCarb Liquid Scintillation Counter, Model 3375. The sample compartment was operated at 8°C. It was noted that 70 to 100 minutes were required for samples to clear after being placed in the cooling compartment.

A total of 56 samples prepared with a tritiated water standard was counted. An average counting efficiency of 27.5 percent was obtained with an average background of 9.5 cpm.



The magnitude of solvent loss from polyethylene vials has been reported [1]. Some loss of toluene resulted after a one-week storage period and some separation of the emulsion occurred. A loss of approximately 20 percent of the original counting efficiency was noted after this length of time [5].

Mention of commercial products used in connection with work reported in this article does not constitute an endorsement by the Environmental Protection Agency.

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NEW PROCEDURES FOR LIQUID SCINTILLATION COUNTING OF TRITIUM USING MULTI-SIZE POLYESTER PLASTIC FILM TUBES

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The growing use of radiochemicals, particularly of tritium-labeled compounds in agricultural, biochemical and medical fields, has produced a need for a simple and inexpensive method of tritium assay. In late 1953, at an early stage of development of the scintillation counter, counting was started in a large 85 ml weighing bottle using 50 ml of the scintillator for each liquid sample of a few milliliters. The counting-efficiency then was 3 to 5% and the background 300 to 400 cpm.

Later, in 1957, with the efficiency increased to 20 to 25% and background lowered to 80 to 100 cpm, counting was changed to the 25 ml size glass vials with 15 ml of scintillator per sample.

Now, with efficiency of 50 to 60%, background of 20 to 25 cpm and the use of extremely high specific-active ^3H-compounds (5 to 100 Ci/mM), it is simple and efficient to count smaller samples into smaller containers with a minimal volume of scintillator.

In 1968, the author [1] published the first preliminary report of a new procedure for liquid scintillation counting in polyester plastic bags to achieve higher efficiency, lower background and less cost than the conventional counting. In 1971, the author [2] reported micro-liquid scintillation counting in polyester plastic minibags with as little as 50 μl of the scintillator per sample, achieving higher efficiency, low background and savings as compared to glass or polyethylene vial countings.

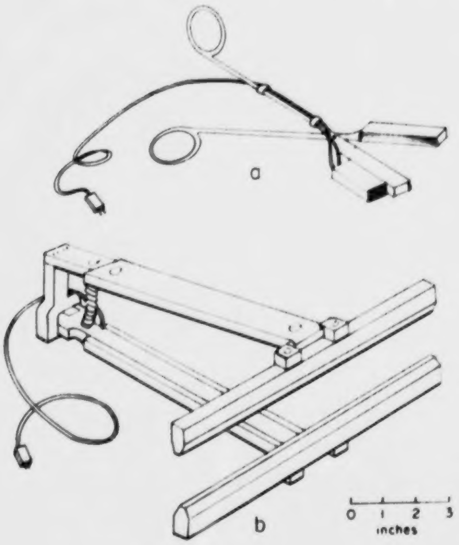
This report describes ultra-micro, micro, semi-micro and standard liquid scintillation counting through the use of multi-size polyester plastic film tubes, which further simplify the counting and accommodate all sizes of liquid samples. Moreover, the tube procedure establishes the trend toward miniaturized scintillation counting that many of us hoped for at the very beginning of scintillation counting almost 20 years ago. Excellent results for other β-isotopes, viz., ^{14}C, ^{32}P, ^{35}S and ^{45}Ca, have been obtained by the tube procedures in this Laboratory, which will be reported elsewhere in the near future. The polyester plastic film used for the tubes is transparent, impermeable, excellently resistant to moisture and to most organic solvents. It is far superior to polyethylene vials in its physical and chemical properties.

Materials and Methods

(a) A small electric heat-sealer (Figure 1-a) with aluminum sealing bars (2" length, 0.3" width and 0.3" depth) is made in the machine shop. (b) A large electric heat-sealer (Figure 1-b) with sealing bars 11" long, 0.5" wide and with a 0.1" sealing edge is also made in the machine shop. (c) Obtain Pyrex glass tubing in 4-foot lengths with 20, 17, 10, 6, and 4mm outer diameter, to prepare tubes in 5 different sizes. (d) Rolls of Scotchpak Brand heat sealable polyester film No. 8, 2 mil thick, are available in varying lengths and widths from 3M Company, St. Paul, MN, 55101. (e) A few hundred wide-mouth counting glass vials and wide-mouth polyethylene vials, used for low background, may be obtained from Demuth Glass Company, Parkersburg, WV. The vials serve as holders for the polyester tubes.

Figure 1

a) Small Electric Heat-Sealer
b) Large Electric Heat-Sealer



The Tri-Carb Liquid Scintillation Spectrometer, Model 3375, was used for all measurements of radioactivity. The efficiency for tritium was 56% in toluene scintillator with 0.5% 2,5-diphenyloxazole (PPO) and 0.01% 1,4-bis-2 (5-phenyloxazolyl) benzene (POPOP).

The polyester plastic film tubes in 5 sizes (Table 1) are made by folding the film around the glass-tubing of a given diameter and sealing the two sides of the film by quickly moving the large heat-sealer (heated to 115°-120°C) from one end to the other. The tubing is cut out from the film with a razor blade or similar device. Several tubes are placed side by side on a flat surface. Each is heat-sealed crosswise with the small heat-sealer (heated to 115°-120°C) at intervals of twice the length of the desired tube size (Table 1). The sealed part is cut in the middle with a pair of scissors to obtain sections of twice the length of the tubes, which are now sealed at both ends. These sections are finally cut in half at an angle, producing tubes of the right length. The angular opening facilitates introduction of the sample and scintillator into the tube (Figure 2).

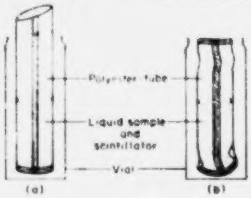
Table 1

Comparison of Size, Scintillator Capacity, Background and Cost Between Tube and Glass Vial Containers

Counting procedure	Tube-size Length X width-I.D. (cm)	Scintillator capacity (ml)	Background (cpm)	Cost/unit (cents)
Ultra-micro	2 X 0.4	0.05	5.0	0.03
Micro	7 X 0.6	0.5	5.0	0.03
Semi-micro	7 X 1.0	2.5	7.0	0.05
Semi-micro	7 X 1.7	5.0	8.0	0.05
Standard	7 X 2.0	15.0	10.0	0.10
Standard	Glass vial	20.0	25.0	15.0

Figure 2

a) Polyester-tube supported in the vial
for introduction of sample and scintillator
b) Sealed-tube held in the vial for counting



With some practice, one to three thousand tubes (depending on size) can be prepared in an hour from polyester film costing approximately one dollar. The tubes are arranged upright in a tray of glass or polyethylene vial holders to be used for the samples.

An aliquot of a liquid sample (organic or aqueous) is pipetted into the right-sized tube, and then the appropriate volume of suitable scintillator. While the tubes are still in a tray of glass or polyethylene vial holders, their upper ends are sealed with the small heat-sealer. Each tube is heat-sealed in a few seconds. The sealed tubes, lying in the holders, are taken to the counter for counting without caps (Figure 2).

Results and Conclusions

Biochemicals which have become available in the last few years have high tritium specific-activities, in the range of 5 to 100 Ci/mM. Hence, an aliquot of a few microliters of the liquid sample has a very low concentration of tritium substrate with sufficient radioactivity. The minimal requirement of scintillator (both solvent and fluor) to count micro-samples for maximum energy transfer and photon yield is at present poorly defined. But the data in Table 2, the results of micro-liquid scintillation counting [2], and other studies [3] provide some direct evidence towards this end. A 1 to 10 µl size sample can be simply counted in an ultra-micro tube with as little as 10 to 50 µl volume of the scintillator (in five to tenfold excess by volume) with similar efficiency and lower background compared to a 25 ml glass vial container with 5 to 10 ml of scintillator.

Table 2

Effect of Scintillator Volume on ^3H Count-Rate in Glass Vial vs. Polyester Tube

		^3H						
		glass vial			tube			
^3H-toluene standard	Scintillator[b] volume (ml)	cpm[c]	effi %	BG[d] cpm	cpm[c]	effi %	BG[d] cpm	Counting procedure
5 µl[a]	10.0	4651	54.4	25	4377	51.2	10.0	standard
"	5.0	4574	53.5	25	4370	51.1	7.0	semi-micro
"	2.0	4369	51.1	25	4420	51.7	7.0	semi-micro
"	0.5	3856	45.1	25	4412	51.6	5.0	micro
"	0.05	3334	39.0	25	4566	53.3	5.0	ultra-micro

[a] 5 µl - 8550 dpm
[b] PPO - 0.5% and POPOP - 0.01% in toluene
[c] mean of triplicates
[d] background

Similarly, counting 50 μl to 500 μl size samples can be successfully accomplished in 0.5 ml to 5 ml size tubes with a much smaller volume (0.2 ml to 4 ml) of the toluene or dioxane based scintillator per sample. The emulsion of gel scintillator [7] will be required in much smaller amounts (three to fivefold in excess by volume) for a given sample. Aqueous samples of as large as 4 ml can now be counted, in a 15 ml tube with 6 ml (40% H_2O) of emulsion or gel scintillator [4,5] with as high as 22 to 25% of ^3H efficiency.

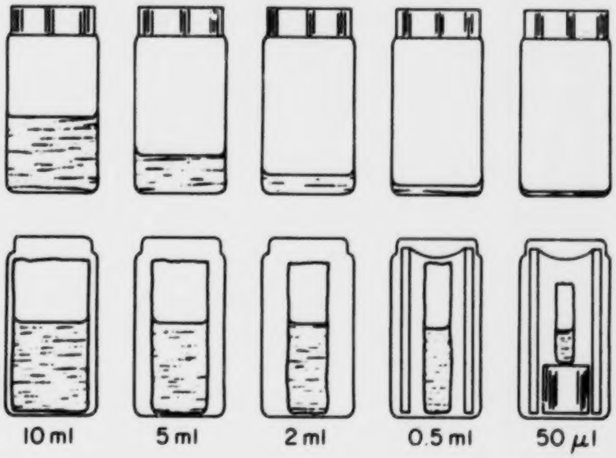
Another factor to consider is the effect of the size of the counting vessel on counting geometry of the machine. Comparative results of counting between the tubes and glass vials are shown in Table 2 and Figure 3. The tritium efficiency in the glass vial decreased gradually from 54.4 to 39% with a decreasing scintillator volume of 10 ml to 50 μl. On the other hand, efficiency in the tubes remains stable at 51 to 53% under similar conditions.

The glass vial is a 25 ml fixed-volume container; therefore, counting geometry diminishes with decreasing volume of scintillator. In the case of tubes, however, it is convenient to keep the scintillator at a constant height, even with as little as 50 μl of scintillator, without affecting the counting geometry. This is another advantage of tubes over the glass vial.

Counting procedures are, thus, simplified through the use of polyester film tubes with the following advantages: (a) heat-sealable, multi-size tubes, easily and cheaply made, (b) ultra-micro, micro, semi-micro and standard countings requiring minimal scintillator, (c) low background and high efficiency, (d) savings over the cost of glass vials and the amount of scintillator, (e) lessening of storage, transportation and disposal problems, (f) minimizing of environmental pollution.

Figure 3

Effect of scintillator volume on the counting geometry for ^3H in glass-vials vs. multi-size polyester tubes



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CHAPTER FOUR
Chemical and Biological Effects of Tritium

RADIATION CHEMISTRY OF WATER
A COMPARISON OF GAMMA RAYS AND TRITIUM BETAS

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The single most important feature of the Radiation Chemistry of condensed systems is the initial nonhomogeneous distribution of reactive intermediates. This distinguishes Radiation Chemistry from other related branches of chemistry, such as photochemistry, and is responsible for the peculiar type of reaction kinetics observed in solution. This initial nonhomogeneous distribution is a direct consequence of the high quantum energies involved, as compared to photochemistry (keV-MeV vs. eV). For example, in the case of high energy gamma rays, energy is deposited initially mainly by Compton scattering. The energetic Compton electron then loses its energy by electronic interactions, causing ionizations and excitations. In water, a secondary electron is usually ejected with sufficient energy to cause several further ionizations and excitations in the immediate vicinity of the original positive ion, leading to the formation of a "spur." A "spur" is a region of high ion density resulting from energy deposition of up to 100 eV. Sometimes the secondary electron has sufficient energy to produce a side-track of spurs of its own. At intermediate energies, these can coalesce into giant spurs (sometimes called "blobs") or short tracks [1]. In general, various sizes of spurs may be formed; the average diameter is a few tens of Angstroms.

These spurs, blobs and short tracks then contain initially positive ions, electrons and excited water molecules. The positive ion is generally considered to have a very short lifetime and rapidly reacts with water to form the OH radical.

$$H_2O + H_2O^+ \longrightarrow H_3O^+ + OH \qquad (1)$$

There have been two major theories on the fate of the electron. One [2] holds that e^- returns to the positive ion to produce an excited water molecule and ultimately H-atom and OH radical.

$$e^- + H_2O^+ \longrightarrow H_2O^{**} \longrightarrow H + OH \qquad (2)$$

The other [3] holds that e^- escapes and becomes solvated.

$$e^- \longrightarrow e^-_{aq} \qquad (3)$$

There is sufficient evidence that hydrated electrons are formed [3,4]. This entity was predicted in the early 50s and demonstrated experimentally in the early 60s. Subsequent reactions of these radicals within the spurs are supposed to give rise to molecular products, H_2 and H_2O_2. These reactions occur while the spur is expanding by diffusion. Those radicals that escape recombination are available to react on ordinary time scales with solutes that may be present in the bulk of the solution. So:

$$H_2O \longrightarrow H,\ OH,\ e^-,\ H_2\ H_2O_2 \qquad (4)$$

This picture of independent yields of radical and molecular species present on chemical time scales was originally proposed [5] largely to account for apparently different qualitative behavior of sparcely ionizing radiations, such as X- or gamma-rays on the one hand, and densely ionizing radiations such as alpha-particles and fission fragments on the other. Early observations on the chemical effects of these radiations seemed to indicate that pure water was not decomposed at all by X-rays where it was decomposed (to H_2 and H_2O_2) by alpha-particles. This was explained on the basis of differences in the initial distribution of the radicals which in turn gave rise to different relative initial yields of molecular and radical products.

Sparcely ionizing radiations have a small value for the linear rate of energy deposition (usually called Linear Energy Transfer, -dE/dX), produce spurs which are initially widely spaced and each have approximately spherical symmetry. Therefore diffusion occurs in three dimensions and escape probability for radicals is high. With higher LET radiations, on the other hand, the spurs are all formed so close together that they overlap into a dense track, diffusion is restricted to two dimensions and escape probability for radicals should be much lower. So at low LET, when diffusion has brought about homogeneity in the system (within about 10 nanoseconds) relatively few radicals will have combined in the spurs and there will be an excess of radicals over molecular products, so the molecular products would be rapidly removed by radical-initiated reactions leading to no net decomposition of the water (no stable products formed). At high LET, where only few radicals escape recombination in the diffusing tracks, molecular products could accumulate to observable concentrations.

These early qualitative studies using radiations of different LET contributed significantly to our present picture of radical products and molecular products existing within about 10 nanoseconds of exposure to the radiation. The fact that the molecular products were formed by radical-radical reactions in regions of high local concentration was demonstrated experimentally by quantitative measurements of the molecular yields in the presence of solutes.

It has been shown experimentally [6,7] that the molecular products are significantly lowered in the presence of a variety of dissolved materials (scavengers). Despite the very low steady-state homogeneous concentrations of radicals (10^{-8} M or less) present in solutions irradiated at the usual dose rates, considerably higher concentrations of scavengers (greater than 10^{-4} M in most cases) were required to significantly lower the molecular yields, even with very reactive solutes. This strongly indicated that the molecular yields are formed in regions of high local radical concentration.

In general is was found that oxidizing agents, which might be expected to react most readily with the reducing radicals e_{aq}^- and H, were most effective in lowering the H_2 yield while reducing agents, which might be expected to react most rapidly with the oxidizing radical, OH, lowered the H_2O_2 yield. Observations such as these indicated that the molecular products, H_2 and H_2O_2, were formed by radical-radical reactions in regions of high local concentrations, two oxidizing radicals giving one molecule of H_2O_2 and two reducing radicals giving one molecule of H_2. These concepts are embodied in the so-called 'diffusions theory' of Radiation Chemistry [7] and several mathematical models incorporating the idea of radical diffusion in competition with radical-radical and radical-solute reactions have been developed, both to explain existing data and to predict new effects [1,8].

Experimental data have been obtained for the radical and molecular yields for radiations of different LET [9]. It has been found that the yield of molecular products increases with LET, as would be expected from the diffusion model. Also, as expected, the hydrated electron yield decreases with increasing LET.

Tritium Results

Within the last two or three years, the basic diffusion model has been theoretically refined in two major directions. The first [1] has involved a more detailed description of the initial spatial distribution of energy deposition by energetic electrons. In this picture, three different types of spatial region are considered, corresponding to different total amounts of energy deposited. The term "spur" is retained for ionizing events in which less than 100 eV of energy is deposited. Between 100 and 500 eV a giant spur called a "blob" is formed, which also has roughly spherical symmetry. When between 500 and 5,000 eV is deposited in an ionizing event, the resulting secondary electron has sufficient energy to escape the parent positive ion, but the resulting spurs are formed so close together that they lose their independent existence and merge into a continuous column. These entities are called "short tracks."

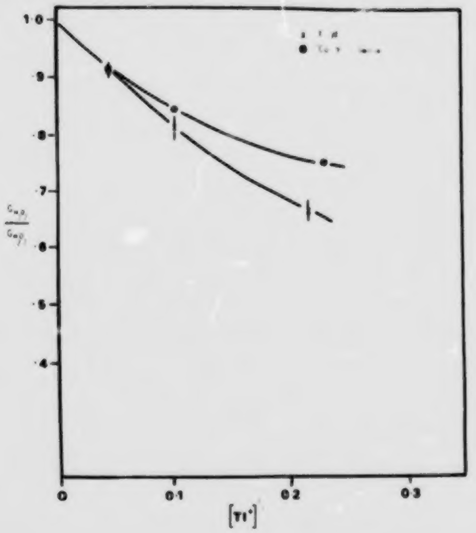
The relative partition of energy among spurs, blobs and short tracks for irradiation by electrons of varying energy is shown in Figure 1. Very energetic electrons, such as Compton electrons produced by gamma radiolysis, deposit their energy mainly in the form of sperical spurs. Lower energy electrons, such as the beta-particles from tritium, show an increasing amount of short track formation. Note, however, that beta emitters give electrons covering a spectrum of energy and, in fact, the average tritium beta-particle has an energy of only 5.6 keV. This model would predict almost entirely short track formation with very little energy appearing as spurs and blobs.

If this description is correct, it should be possible to compare the chemical properties of short tracks and spurs by parallel experiments using either gamma-rays or tritium beta-particles. In this case, the yield of H_2 or H_2O_2 should be somewhat more difficult to suppress by added solutes if it were formed in short tracks rather than in spurs, because the reactive intermediates, the radicals, are formed in much closer intitial proximity.

To test this hypothesis, the H_2O_2 yield was measured as a function of both Br^- and Tl^+ as scavenger. These ions are known to react

Figure 3

Decrease in $G_{H_2O_2}/G^o_{H_2O_2}$ with concentration of thallous ion in 0.4M H_2SO_4: ⊙ tritium β radiolysis; ●, γ radiolysis [12].



rapidly [10] with OH radicals, the supposed precursors of H_2O_2, and hence suppress the H_2O_2 yield. It was found that in both cases the H_2O_2 yield in tritium beta radiolysis was more easily suppressed than in the case of ^{60}Co-gamma radiolysis. This can be seen in Figures 2 and 3 which show the relative lowering of $G_{H_2O_2}$ as a function of Br^- (in neutral solution) and Tl^+ (in 0.8N H_2SO_4) concentration, respectively, for tritium beta radiolysis. Literature data for gamma radiolysis [11,12] is also shown (the cube root of the solute concentrations is used to spread the data). $G^0_{H_2O_2}$ is the value extrapolated to zero solute concentration. Similar differences in scavenging of H_2O_2 between γ and tritium-β radiolysis have been reported [13] for Cl^- and Br^- in 0.8N H_2SO_4.

A possible explanation might involve the longer time scale over which H_2O_2 is formed in short track geometry as compared with spur geometry. This longer time scale arises because the short tracks can be considered as regions where a number of spurs are formed initially close together. As each spur expands, inter-spur reactions would lead to H_2O_2 formation on a relatively long time scale in addition to the intra-spur reactions. Hence, in the case of tritium beta radiolysis, scavengers would have a longer time scale to react with the radical precursors of H_2O_2 than in the case of gamma radiolysis, so their effect would be more pronounced, as is seen in Figures 2 and 3.

In the case of the molecular hydrogen yield, the situation is not so simple. It appears probable that there are several processes which lead to the formation of hydrogen in radiation chemistry. Recent calculations [14] based on the diffusion model show that, in order to explain scavenging experiments using gamma-rays it is necessary to build into the model four different sources of H_2 at neutral pH. With their relative contributions they are

$$e^-_{aq} + e^-_{aq} + 2H_2O \longrightarrow H_2 + 2OH^- \quad (31.6\%) \quad (5)$$

$$e^-_{aq} + H + H_2O \longrightarrow H_2 + OH^- \quad (29.7\%) \quad (6)$$

$$H + H \longrightarrow H_2 \quad (\ 3.9\%) \quad (7)$$

together with an "initial yield" of H_2 (34.8%) [14]. All these processes occur in the spur during its diffusion. There is no reason to suppose that the relative contributions of these processes are invariant with LET. In fact, the relative number of H atoms to e^-_{aq} escaping the spur has been shown [9] to increase as LET increases. The effect of scavengers in the case of molecular hydrogen would then be complicated by changes in the nature of the important precursors, as well as in their spatial distribution. We are currently performing experiments on scavenging of G_{H_2} designed to test these factors.

In summary, there is both theoretical and experimental evidence that tritium betas deposit their energy in the form of short tracks of high local LET. In view of the well-known importance of LET in biological systems, further evidence for short track effects in tritium beta radiolysis might be found if appropriate biological systems are investigated.

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SOMATIC AND TERATOGENIC EFFECTS OF TRITIUM

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There are numerous sources of tritium, all of which have been amply discussed in this book. When tritium is uniformly distributed throughout the body, there is no reason to believe that it will produce in body water, qualitatively different effects than an equivalent amount of radiation of any other source. Quantitatively, an appropriate quality factor must be used [1]. However, there has been considerable apprehension about the incorporation of tritium into genetic material as the result of the very short range of the tritium beta-particle and the possible effects of transmutation. In principle, one must consider the question of chemical transmutation, nuclear recoil and changes of charge producing positive ions. The general problem has been comprehensively reviewed by Bond and Feinendegen [2] in respect to the hazards of intranuclear tritiated thymidine. The important conclusions from their analytical review of data are:

1. "The absorbed dose concept holds down to the order of 10^{11} gram or less, and somatic effects can be predicted on this basis.

2. "The distribution of tritium atoms incorporated into DNA as tritiated thymidine (and ion pairs from the beta particles) are randomly distributed as far as dose calculations for the purpose of predicting somatic effects are concerned.

3. "Every part of the cell nucleus lies within one tritium beta range of some part of a chromatid, and the nucleus contains no sizable contiguous insensitive volume of a radius exceeding the effective range of tritium beta rays (1 to 2 μm).

4. "The origin of tritium beta tracks in, or their close juxtaposition to, the DNA molecule does not appear to enhance the degree of somatic effects."

In the study [2] based on all data published at the time, the degree of early somatic biological effect from intranuclear tritium is that anticipated on the basis of calculated averaged absorbed dose to the cell nucleus, at least down to doses of 4 rads, corresponding to about 15 tritium beta events in the nucleus.

Tritium Dosimetry

Tritium

The half-life of tritium is 12.3 years; it decays to helium-3 by emitting beta-particles with a mean energy of 5.7 keV and maximum energy of 18.6 keV. The mean range of the beta-particle in water is 0.8 μm and the maximum range approaches 6 μm.

The tritium beta has generally been considered to have a quality factor (Q) close to unity [2]. From the discussion and observations of Johnson [1] presented elsewhere in this book, it is suggested that a more appropriate Q is 2 to estimate biological risks from a given dose of tritium beta radiation.

In the context of this paper, the possible hazards of tritium in the form of tritiated water and tritiated thymidine will be considered. If tritium is uniformly distributed throughout tissue, the usual calculation of dose from radioisotopes applies except at very low concentrations, as will be discussed later. One μCi of tritium distributed uniformly per gram of substance of unit density delivers a dose rate of 12.14 millirads per hour. Hence, knowing the concentration of tritium in tissue and its biological half-life of roughly 10 days in man, the total dose to be delivered from any concentration of tritium is easily calculated by standard principles covered in Handbook 69 [3] and presented by Johnson [1].

Another factor of considerable importance in considering the toxicity of tritiated water is the content of water in soft tissues. The gross concentration of water is tissues varies between 0.6 and 0.8. However, on the basis of centrifugal studies, it is clear that there are different densities of nuclei and cytoplasm. Since the range of tritium beta-particles is very short, the concentration of water within nuclei may be very important. The average concentration of water in tissues probably does not represent concentration of water in nuclei, see Johnson [1]. All of the preceding considerations must be taken into account when calculating dose of radiation from any constant level of tritium in the form of water and following a single dose of tritiated water.

The biological half-time of water may vary considerably, depending upon water and electrolyte intake and hormonal influences. The turnover rate of water has been studied in many animals including man. In patients, Rubini *et al.* [4] found the half-time of water to vary from 7.25 to 12 days, presumably dependent upon water intake. In long-term studies on exposure and subsequent turnover of tritium in the form of water, the excretion pattern may be regarded as the sum of exponentials with half-times of disappearance of approximately 6, 23 and 345 days [5-9]. There are several other discussions on tritium turnover under various conditions in man in this book.

Somatic Effects of Tritiated Water

Studies on lethality of tritium were initially performed by Brues *et al.* [10] who found the $LD_{50/30}$ of tritiated water for mice to be approximately 1 mCi/g body weight administered in a single injection. The anatomic modes of death were similar to that observed after whole body irradiation. The accumulated dose during the exponential decrease of body water as a result of turnover of water is calculated to be in the vicinity of 700 rads. The actual dose is dependent upon tissue concentration of water and the actual rate of water turnover, all of which have an error of estimate.

Splenic and Thymic Atrophy

Storer *et al.* [11] maintained relatively constant levels of tritiated water in body water for five days and then measured the atrophy of mouse spleen and thymus along with the decrease in ^{59}Fe uptake in the red cells of rats. The atrophy observed and depression in ^{59}Fe uptake was somewhat more than one would expect from a comparable dose of X-ray given over the same period of time leading to an estimate of RBE between one and two.

The Effects of Tritiated Water on the Embryo

Cahill and Yuile [12] maintained pregnant rats at constant body activities of tritiated water ranging from 1 to 100 μCi/ml of body water throughout pregnancy. The range in dose rate from these concentrations of tritium in body water to the embryo and fetus was approximately 0.3 to 30 rads per day. The only detectable changes in offspring exposed to 1 μCi/ml during pregnancy were slight differences in liver and heart weight. At higher levels, there were statistically significant findings including microencephaly, sterility, stunting, litter size reduction and increased resorption of the fetuses. The decreases in the absolute and relative weights of the fetal organs were roughly proportionate to dose of radiation. Histologically, all of the organs with the exception of gonads were approximately normal. During a 270-day observation period after birth, five tumors occurred in the irradiated animals compared to one in a control animal.

Hatch *et al.* [13] have studied kangaroo rats living in a tritiated environment surrounding the crater of a nuclear detonation in Nevada. The tritium activity of body water in tissues of these animals ranged from 0.1 to 0.4 μCi/g of body weight. Organically bound tissue tritium had approximately the same specific activity as body water. It appeared as if the animals were in equilibrium with their food supply and the soil so far as tritium concentration was concerned. The lifetime radiation dose to the adult animals was estimated to be 10 rads from internal tritium and 10 rads from the beta- and gamma-emitters in the soil. The authors found no gross evidence of harmful radiation effects in these kangaroo rats. The animals grew normally and reproduced well in the vicinity of the crater. Admittedly, there is not a great deal of information on the natural incidence of disease, longevity and incidence of tumors in this wild strain of rats.

Although further data are required to set the levels at which barely detectable harmful somatic effects from tritium will be found, it can be "guesstimated" that it is in the vicinity of 0.5 to 1 μCi/g of body weight in small mammals. It is probable that the permissible level will be set by the level at which one finds no detectable genetic effects.

The Effects of Tritium on Killing of Spermatogonia in the Mouse

The comparative effect of tritiated water and external X-rays was studied by Lambert [13] 72 hours after the intraperitoneal injection of 20 μCi/g of body weight of tritiated water, the effect on primary resting spermatocytes was equivalent to the effect of 30 rads of 200 kVp X-ray. The RBE is these studies was 2.4. Since the technique is able to detect about 5 rads exposure to external radiation, one could predict that the amount of tritium which would give an integrated dose of about 2 rads would be detectable.

Tritiated Thymidine

There has been considerable concern about tritiated thymidine (^{3}HTdR), since it is a specific DNA precursor. In man it has been shown that 40 to 60 percent of an injected dose of ^{3}HTdR is catabolized to tritiated water and the remainder is presumably incorporated into DNA [4]. Only those cells that are in the DNA synthesis phase of the proliferative cycle incorporate ^{3}HTdR, hence there is a cohort of labeled cells. This introduces a marked inhomogeneity in the radiation exposure of cells; some receive no radiation, and others receive a wide range, depending on the amount of ^{3}HTdR incorporated. In order to study the dose delivered to the genetic material of chromosomes from tritium incorporated into bone marrow cell nuclei, the notion of an average nucleus as a sphere of 8 μm and a volume of 270 X 10^{-12} ml(g) was introduced [2]. From the range dose relationship for tritium beta rays, approximately 20% of the energy will escape from the average nucleus. This notion has been extended to the idea of a "standard nucleus" [16] for evaluation of hazard from intranuclear radiation in general. The beta rays have a maximum range in water of approximately 6 μm and an average range of 0.8 of 1 μm. With spheres smaller than the "standard nucleus," an increasing amount of radiation escapes from the nucleus, and in spheres larger, an increasing amount is retained within the nucleus. For spheres of 12 μm in diameter, nearly all of the energy will be deposited within the sphere [15]. From the average energy of the tritium beta, it can be shown that there is 0.37 rad if all of the energy of a tritium disintegration is deposited in a mass equal to the "standard nucleus" of 270 pg [2,16]. However, as mentioned earlier, 20% of the radiation escapes; hence, for each disintegration within the "standard nucleus," there is an absorbed dose of 0.3 rad. In the case of TOH at relatively high concentrations, the amount of radiation escaping from the nucleus will be neutralized by an equivalent amount of radiation entering the nucleus from the cytoplasm.

Intranuclear Tritium Irradiation in Tissue Culture

The cell killing effects of tritiated thymidine have been analytically reviewed [2]. When all factors are taken into consideration, the effect upon dose effect curves are comparable to what would be expected from the intranuclear tritium dose to the cells that have incorporated ^{3}HTdR.

Cytogenetic effects (chromatid and chomosome type aberrations) have been observed by many investigators and are also reviewed [2]. In general, the effects observed could be predicted from the radiation dose to the cells as calculated on the basis of the number of tritium disintegrations that had occurred in the cell.

Radiation Effects of Tritiated Thymidine in the Intact Mammal

Feinendegen [17] has determined the smallest amount of intravenously injected ^{3}HTdR that would result in a detectable effect in the bone marrow of the rat. The end point was a change in the turnover rate of labeled bone marrow cells by determining radiochemically and autoradiographically the amount of radioactivity per unit volume of bone marrow suspension. After 1 μCi/g ^{3}HTdR, a definite diminution in the turnover rate was observed. A questionable change was observed at 0.5 μCi and no change was observed at 0.1 μCi of ^{3}HTdR/g body weight.

The dose distribution to the labeled bone marrow cells was determined on the basis of the distribution of grains overlying the labeled cells, and the energy deposition from the tritium betas. The effects observed could be predicted on the basis of anticipated radiation injury from the calculated doses to the more highly labeled cells.

"Thymidine suicide" is a technique that has been used for the study of bone marrow stem cells. Smith *et al.*[18] found a significant loss of proliferative capacity of bone marrow cells exposed *in vivo* to four doses of 10 μCi/g of ^{3}HTdR over a 40-hour period. Bruce and Meeker [19] observed a loss of colony-forming capacity of cells in the marrow or spleen of mice exposed to six doses of 1 mCi of ^{3}HTdR given *in vivo* over a 24-hour period. Becker *et al.* [20] have incubated bone marrow suspension for 20 minutes to large amounts of ^{3}HTdR. A maximal killing of cells in DNA synthesis is attained at about 150 μCi/ml of incubation medium.

The influence of ^{3}HTdR on lymphocytes has been studied by giving graded doses of ^{3}HTdR to rats and killing the animals five hours after the intravenous administration [21]. The end point used was that of pyknosis of lymphocytes developed by Trowell [22]. No effect was observed after 5 μCi of ^{3}HTdR specific activity 1.9 Ci/mM. At higher doses, an apparent linear effect was observed up to 25 μCi/g of rat. By comparison to the percent pyknosis produced by whole body X-ray, 25 μCi is comparable to 75 rads.

Greenberg *et al.* [25] have observed striking cell killing effects in a leukemic patient given 0.25 μCi/g body weight in each of 10 injections over a five-day period. Effect on spermatogonia have been studied by Johnson and Cronkite [23] and Kisieleski *et al.* [24]. The latter has observed questionable effects at 1 μCi/g of injected ^{3}HTdR. There were significant effects at 10 and 50 μCi/g. Johnson and Cronkite [23] estimated that 1 μCi/g delivered an averaged dose rate of about 2 rads per day during the period between injection and killing of the animals, producing an effect comparable to about 5 rads protracted over this period of time.

Grisham [26] has studied the influence of ^{3}HTdR upon the regeneration of partially hepatectomized rats after doses of 0.25 to 5.0 μCi/g of body weight. At doses of 1 μCi/g body weight and greater, a transient inhibition of regeneration is induced as measured by the rate of increase of residual liver mass, mitotic rate and the incidence of abnormal mitotic figures.

Carcinogenic Effects

There have been three distinct studies in which the carcinogenic effect of ^{3}HTdR was investigated following injection into mice. In the first study, Cottier et al. [27] injected 10 μCi/g body weight, specific activity 1.9 Ci/mM at weekly intervals for three weeks into C-57Bl/6J mice. Whole-body irradiation was used as a control. There was a significant induction of tumors as expected from a total of 480 rads whole-body irradiation given in increments of 160 r at seven day intervals. The ^{3}HTdR treated animals did not develop any thymic lymphomas. Baserga et al. [28,29] have administered thymidine in doses of 0.1 to 10 μCi/g body weight into embryonic, newborn and mice up to 14 months of age. The higher the dose of ^{3}HTdR and the earlier it was given in life, the higher was the incidence of tumors compared to the control mice receiving no radiation in any form. Johnson and Cronkite [30] injected 1 and 5 μCi of ^{3}HTdR/g of body weight into Swiss Albino mice at 6 to 12 weeks of age. The results in these mice were compared to non-irradiated controls and animals receiving 400 r of whole body gamma radiation. The mean life spans and age specific mortality rates of mice injected with ^{3}HTdR were indistinguishable from those of the control animals. There was no measurable alteration in tumor incidence, tumor type or time of onset of neoplasia. The differences in response between the investigators may be due to a difference in the strain susceptibility of the mice. One can tentatively set a dose of 1 μCi/g body weight as probably the lower limit at which an increase in tumor incidence may be detectable in mice.

Effects of ^{3}HTdR on Oocytes

Haas and Fliedner [31] have administered ^{3}HTdR to pregnant rats from the ninth day of pregnancy until parturition. At intermittent intervals from 1 to 240 days after parturition, the treated animals were killed and the ovaries compared to those of untreated controls of the same age. The pregnant animals were given continuous intravenous infusions of ^{3}HTdR that delivered 290 μCi/day to the pregnant animals. There was a striking diminution in the number of primary follicles and the later development of Graafian follicles.

Effect of ^{3}HTdR on Embryos

Chicken embryos at 60 to 69 hours of age were given 15 μCi of ^{3}HTdR. Control embryos were given 200 r of X-ray. Half of the ^{3}HTdR treated chicks were grossly normal, the other half showed a slow development and hatching with neural and muscular defects of the hind limbs. The ^{3}HTdR treated chicks had less effect than those given 200 r of X-ray.

Fliedner et al. [32,33] have studied the effects of continuous administration of various doses of ^{3}HTdR to pregnant rats from 1.6 μCi/g-d to 8 μCi/g-d. At 1.5 μCi/g-d, there were no obvious injuries to the pregnant rats nor were there any impairment in the general or hematopoietic development of the offspring. At doses greater than 1.6 μCi/g-d there was a marked bone marrow depression in the pregnant animals and an increasing frequency of stillborn rats. The neonatal growth as well as the development of the blood forming tissues was impaired the higher the dose of ^{3}HTdR had been to the mother. Rats given 8 μCi/g-d had marked malformation in the fetus including abnormal development of several organ systems.

General Discussion of Effects of ^{3}HTdR in the Mammal

Below an injected dose of 1 μCi/g body weight there have been practically no detectable effects of ^{3}HTdR. Johnson [34] has pointed out that in a wide series of studies, effects are detected at doses greater than 1 μCi/g body weight for tumor induction [28,29], killing of mouse spermatogonia [30], diminution and turnover rate of rat bone marrow cells [2], pyknosis of the lymphocyte [21], inhibition of liver regeneration in the rat [26], and lastly, the induction of hepatocyte polyploidy in the rat [35]. From autoradiographic studies on spermatogonia [30] and in rat bone marrow cells [2], it has been estimated that the dose rate is 2 to 8 rads per day for these tissues, respectively. However, almost all cells in the mammal that become labeled have relatively short cell cycles, from 12 to 48 hours. Admittedly, some cells may have much longer cell cycles and thus accumulate higher doses of radiation as the cell cycle may be much longer in "stem cells" and account for carcinogenic effect.

When ^{3}HTdR is presented to an animal only in a single pulse, this cohort of cells will dilute out the intensity of label in the chromosomes by replication from nonradioactive materials at the next DNA synthesis. As pointed out by Cronkite et al. [36], it can be expected that the labeled chromosomes will be randomly distributed among the daughter cells at each mitosis. In human cells with 46 chromosomes, about five percent of the cells will have zero labeled chromosomes after the fifth division. After the sixth division the most frequent number of labeled chromosomes is one and 23% of the progeny can be expected to have no labeled chromosomes at all. In addition, another feature of somatic cell proliferation is the fact that a substantial portion of the progeny mature into nonproliferating cells which differentiate and die or are lost from the body, thus removing the labeled DNA.

From the preceding review of the effects of TOH and ^{3}HTdR after high single, multiple or chronic exposure, it is quite clear that one can produce major radiation effects in the mammal. Where there are sufficient data to estimate the dose of radiation and using an appropriate Q, the effect observed is comparable to what one would expect from external radiation. The major question is whether effects are linear down to background levels of radiation or whether, at very low doses of internal or external radiation, some cells receive no radiation. Lastly, can the cells exposed to tiny doses essentially repair all of the injury? From the presentation of Russell [37], repair of genetic injury is evident. After radiation exposure, it is generally believed that single strand breaks are usually repaired and double strand breaks are usually irreparable, leaving residual injury. Let us examine some of the factors concerned with very low level exposure and the probability that there will be one cluster of ionizations or "absorption events" in a volume equal to the "standard nucleus."

Very Low Level Exposure

When considering very low levels of TOH, intranuclear radiation or background levels, one becomes concerned with the energy absorbed per volume of cell nuclei containing the genetic material (chromosomes), which is believed to be the radiosensitive site in the cell. As mentioned earlier, mammalian cell nuclei vary considerably in size. The "standard nucleus" has been defined as a sphere of 8 μm diameter with a volume of 270 X 10^{-12} ml(g) in order to calculate dose to the nucleus when the low energy beta-emission of tritium occurs within the nucleus. On an average 20% of the energy will escape from the "standard nucleus."

The absorbed dose is expressed as rads (100 erg/g tissue). One can relate ergs to ionizations per unit volume of tissue. The ionizations are formed in clusters over the path length of a charged particle and not randomly. Path lengths vary with energy. Such clusters can be called "absorption events" and may supplant the rad at very low levels of exposure when small volumes of tissue may be receiving no exposure over long intervals.

First let us consider the probability that a "standard nucleus" will be hit when exposed to 0.1 rad of 100 keV X-ray photons:

$$0.1 \text{ rad} = 10 \text{ ergs/g} = 6.242 \times 10^{12} \text{ eV/g}$$

$$= 6.242 \text{ eV}/\mu\text{m}^3 = 0.1836 \text{ ion pairs}/\mu\text{m}^3$$

In a volume of the "standard nucleus" of 270 μm^3 there will be 270 X 0.1836 or 49.57 ion pairs per cell nucleus on the average. Therefore, if ion pairs were randomly distributed, all cell nuclei would be hit. However, ion pairs are bunched in delta ray tracks with ranges up to 3 μm.

Because of this, ionizations may be produced in the nucleus from primary events occurring up to 3 μm beyond the nuclear membrane. Adding 3 μm in all directions to the 8 μm diameter of the standard nucleus gives a sphere of 14 μm diameter and 1437 μm^3 volume, within which primary events that can affect the nucleus occur. When a primary event occurs beyond this boundary, the delta rays will not reach the nucleus. The 1437 μm^3 sphere receives 0.1836 X 1437 = 264 ion pairs.

Since a 100 keV photon loses about 0.14 of its energy per primary event, there are $\frac{100000 \times 0.14}{34} = 412$ ion pairs per track. Thus the 264 ion pairs produced in the 14 μm diameter sphere represent an average of 264/412 = 0.640 tracks per 14 μm sphere. All of these originating in the nucleus will be effective, but only something of the order of, but less than, half of those originating in the surrounding 3 μm thick shell of cytoplasm will reach the nucleus. For simplicity, assume that a total of 150 ion pairs affect an average nucleus (50 from tracks originating in the nucleus plus 100 from the 3 μm shell). This reduces the number of effective primary events to 150/412 = 0.364 per nucleus. Assuming a Poisson spatial distribution of these events, $e^{-0.364} = 0.695$

is the fraction of cell nuclei which receive "zero" dose.

Comparable analysis of natural background would be of interest but the wide spectrum of energy and type of the external and internal components of natural background complicates the analysis. However, it is clear that in considering low level exposure to external radiation in vicinity of 0.1 rad of 100 keV X-rays many "standard nuclei" receive no radiation exposure.

A similar analysis for TOH follows:

$$1 \text{ rad} = 100 \text{ erg/g}$$

$$100 \text{ erg/g} = 62.4 \times 10^{6} \text{ MeV/g}.$$

Since 34 eV are transferred per ionization, there are 1.83×10^{12} ionizations within a gram per rad absorbed. These ionizations are grouped into clusters the size of which is dependent upon the energy of the β particle. With tritium beta there are an average of 180 ionizations per "absorption event." Thus, for each rad absorbed from tritium betas, there are about 1.0×10^{10} "absorption events" per rad. Hence with random distribution of tritium in tissue, there are 0.01 "absorption events" per μm^3 of tissue per rad absorbed or 2.70 "absorption events" for each volume equivalent to a "standard nucleus." To deliver the maximum permissible dose of 5 rem per year would require 13 to 14 tritium beta emissions per year. The maximum permissible chronic body burden of 1 mCi/ml of body water gives a weekly dose of 37 mrad or 1.9 rem per year.

With a body burden of 0.1 mCi tritium there is 0.0023 μCi/ml of body water. There will be 2.69×10^{9} disintegrations per year per ml or 2.69×10^{-3} $d/\mu m^3$-a. There will be on an average 0.73 $d/270\ \mu m^3$-a the volume of the "standard nucleus." Assuming a Poisson spatial distribution, $e^{-0.73} = 0.482$ is the fraction of "standard nuclei" that receive "zero" dose. A similar calculation for 1 mCi body burden shows that 0.0007 nuclei receive zero dose. Accordingly as concentrations of tritium decrease below the chronic maximum permissible body burden of 1 mCi/standard man, there will be an increasing fraction of cells that receive no radiation from TOH. The cell nuclei in which a decay occurs will generally receive only one absorption event/year or an average dose of about 0.3 rad since at these low concentrations the loss from the nucleus will not be neutralized by irradiation from the cytoplasm.

All experimental data indicate recognizable biological effects following an acute body burden of about 1 μCi ^{3}HTdR/g of body weight. This leads to about 1.5 disintegrations per hour or one absorption event per 40 minutes to the average labeled bone marrow cell in rat and man [2,16]. The dose rates to the single cells range probably from one absorption event every 10 minutes to one absorption event every 160 minutes for the lightly labeled cells. In terms of rads, this amounts to an average of 0.3 rad per 40 minutes to the labeled nuclei with ranges from 0.3 rad per 10 minutes to 0.3 rad per 160 minutes. Concerning the single labeled cell, it is meaningless to express the dose in terms of a fraction of 0.3 rad, despite the fact that a single tritium decay in the nucleus has a small probability of irradiating predominantly the cytoplasm.

It is reasonable to consider that the radiation effects in the mammal are a function of the product of the number of cells involved and the average dose absorbed per involved cell. At levels of tritium below the maximum permissible concentration, with rare statistical exceptions, all involved cells will receive only one "absorption event" or 0.3 rad/year. The probability that the same cells will be involved each year is remote. The fraction of cells involved will decrease with tritium concentration. At 1/7 of maximum permissible concentration, 1 of 10 cells would be involved/year and it would take more than 10 years for all cells to receive at least one "absorption event" of 0.3 rad.

Although there are no experimental studies at low levels of radiation, it is interesting to speculate about the possible role of neighboring uninjured cells in helping repair injury of involved cells. Even though it may be nearly impossible to study such a proposed mechanism, it is not unreasonable to propose it. There are ample data showing that shielding of bone marrow and transplantation of marrow significantly reduces the incidence of leukemia in irradiated mice.

At this low level of exposure, there is a long time for repair mechanisms to operate. Veatch and Okada [38] have shown that in one mammalian cell line, mouse leukemia (L5178^{Y}), there are three single strand breaks/cell (G_1 stage) per rad and 0.3 double strand breaks per cell (G_1 stage) per rad. Single strand breaks can be assumed to to be repaired. The double strand break is usually considered irreparable. Since one tritium "absorption event" represents 0.3 rad, there will be about one single strand and 0.1 double strand breaks, hence only one of ten involved cells will retain residual injury. This, along with the decreasing number of cells involved with concentrations of tritium below the maximum permissible concentration, leads one to speculate that the slope of the radiation effect curve obtained at high radiation levels, where it is easy to observe effects, may not be applicable to the very low levels where many cells are spared and there is a long time for repair of the injured fraction of cells.

ACKNOWLEDGMENTS

Research leading to the production of this paper was supported by the U. S. Atomic Energy Commission.

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THE BIOLOGICAL EFFECT OF CERTAIN TRITIUM LABELED COMPOUNDS RELATED TO DOSE

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In radiological protection, tritium has traditionally been considered relatively "safe." This is due partly to the low energy of its β-emission (about 1 μm in water), resulting in virtually no external dose, and also because of the mass of information which has accumulated concerning its most common compound, tritiated water [1-8]. The Recommendations of the International Commission on Radiological Protection Committee II [9] contain data on this compound and tritium gas only; it has been shown [10] that the maximum permissible annual intake of tritiated water is about 60 mCi. This amount of activity appears relatively large compared with other radionuclides, but the sensitivity of modern detection and counting systems makes it rarely necessary to use such large amounts of activity in research. Thus, the greatest risk in experimentation with tritium is that of cross contamination rather than contamination of personnel. However, tritium has always been potentially hazardous; being an isotope of hydrogen, it may be incorporated into biological active materials and thence into human tissues. This potential was realized quickly following the introduction of tritium-labeled biologically active compounds, in particular tritiated thymidine [11]. Tritiated thymidine reaching the blood may be incorporated into the DNA of cells during the synthesis stage prior to mitosis. In this way tritium may be concentrated in the nucleus, which is the most radiosensitive part of the cell, and thence irradiate this structure almost exclusively. Early theoretical work indicated that tritiated thymidine should be considered several orders of magnitude more toxic to mammalian cell systems than tritiated water. Although later work has disproved this concept, there are still reports published suggesting that, on the basis of dose calculation, tritiated thymidine should be considered an extremely hazardous material. For example, Guild [11] concluded theoretically that "tritiated thymidine can deliver a dose to the chromosomes of radiosensitive cells from 50 to 50,000 times greater than that delivered by an equal activity of tritiated water." Oliver et al. [12], with reference to in vitro work, emphasized the care with which tritiated thymidine should be handled. Later work, notably that of Dewey et al. [13], using chromosome aberrations, indicates that intracellular tritiated water and tritiated thymidine are about equally effective in producing cytological damage. Lambert [14], using the system described later has shown that, in vivo, tritiated thymidine is only about four times as effective in killing cells as tritiated water when related to injected activity. However, two authors have concluded that in one instance "tritiated thymidine is more hazardous as an internal emitter than tritiated inorganic molecules such as water or hydrogen gas by at least a factor of 20," [15] and also that "the MPCs for tritiated thymidine should be about 10^{-4} to 10^{-5} times the MPCs for tritiated water" [16]. In common with most radioactive materials, there are very little data on the relationship between radiation dose and biological effect in humans, but there have been a number of animal experiments designed to evaluate this relationship for tritiated thymidine [14,17].

Tritiated thymidine is a special case because it is incorporated into DNA and therefore should be more hazardous than other compounds. Conversely, it is selectively incorporated by rapidly proliferating tissues which, although they are generally more radiosensitive, can sustain considerable cell depletion without loss of ability to survive or regenerate to full capacity. In fact, natural cell loss is an essential feature of such organs and tissues, e.g., the bone marrow, gut lining, etc. There are many more compounds which may be incorporated into cell constitutents, e.g., uridine (principally into RNA), amino acids (into protein), etc., and there is a paucity of data of radiobiological effect on such compounds. Because of this general lack of relevant data, a selection of compounds, which is taken up into different parts of the cell and therefore irradiate different volumes within the cell, was included in this study. Previous work carried out here [8,18] has attempted to evaluate dose to organs but more recently [14] to relate dose to subcellular volume (the nucleus) to effect produced. This was largely unsuccessful because of the enormous difficulties of determining, in vivo, the actual amount of tritium within a nucleus which initiated the death of a cell. It is also difficult to compare effects due to irradiation by tritiated water, which is essentially uniform throughout a cell population, to effects seen after administration of tritiated thymidine, which produces a heterogeneous dose pattern - some cells being only irradiated by the tritiated water of catabolism. However, from this work it has been shown that tritiated thymidine is no more than a few times more toxic to cells than tritiated water.

Normally, the radiation dose is measured at the point at which the biological damage is observed. Although in this case cell death was observed, it is not altogether clear over which volume within the cell the dose should be averaged; i.e., should the dose to minute volumes of the nucleus be estimated: dose to the chromosomes of dividing cells; the DNA as a whole; the nucleus; the labeled cells or the whole organ? This sort of problem is important because it has been calculated [12], for example, that the decay of a tritium atom within the nucleus produces a dose of about 170 rad to a sphere of tissue 1 μm in diameter, a dose large enough to severely damage the cell if located in a radiosensitive area. Thus, the work reported here attempts to investigate whether biological effect can be predicted solely from a knowledge of the average organ dose.

Cytological Features of the Testes of Relevance to this Work

In general, the problems of measurement of biological effect and dosimetry are far more difficult in vivo than in vitro mainly because the population of cells is not homogeneous in respect to their size and rate of replication. For this reason, the greatest volume of literature concerning radiobiological effects involves experiments on cells in culture. Nevertheless, it is usually difficult to extrapolate such results to the complex cell systems of human tissues. However, of all the organs within the body, the testes offer some possibility of making quantitative measurements of radiation damage. The fact that the testes are an isolated cell system, essentially outside the body and losing only mature cells after a well-defined division and maturation cycle together with the easy recognition of cells, facilitates detection of responses which might not be observed in other systems. Because of this and the relative radiosensitivity of some species of germ cells [19] a considerable amount of literature concerned with irradiation of the testes has been published. Thus, the cellular kinetics of the seminiferous epithelium are well known for several animal species, in particular the mouse. For these reasons, cell death in the mouse testes has been chosen as the criteria of radiation damage for several studies made here. As the timing of spermatogenesis is complex and important to this experiment, it is described in some detail here.

The mature mouse testis consists of two main components: convoluted seminiferous tubules and interstitial tissue lying among them. The seminiferous tubules contain the sustentacular cells of Sertoli and spermatogenic germ cells which, by proliferation and complex morphological transformation, develop into mature spermatozoa. Adult testes contain a mixed population of cells at all stages of development. In any histological section, four main cell types are present in different states of maturation. The layer of cells nearest the basement membrane of the tubule consists of the most immature, namely spermatogonia, in some phase of their development. Cells derived from these, primary spermatocytes, occupy a layer slightly nearer the lumen. The next layer consists of either spermatocytes in meiotic division or spermatids. The fourth layer of cells, nearest the lumen, is made up of numbers of maturing spermatozoa "spiralling" towards the center. Thus, in a tubule cross section there will be the same combination of cell types around its periphery. However, cell combinations vary continuously along the length of the tubule, certain combinations occurring at regular intervals.

The mouse testes have been studied extensively [20-25] and these studies have resulted in a method of classification of the various stages of development of cells in the seminiferous epithelium. The method used depends upon the classification of the development of the seminiferous epithelium into twelve states (normally denoted by Roman numerals) on the basis of the changes in morphology of the nuclear cap or acrosomic system of the heads of the spermatids as revealed when stained by the periodic acid-Schiff (PAS) technique [26]. This classification is then used to define the concomitant development of the spermatogonia and spermatocytes. Details of the lengths of the intermediate stages of spermatogenesis given by Oakberg [21] have been used in this work.

It has been shown that type A (or stem cell) spermatogonia undergo several divisions at tubule stages IX, XI and II. After the third division, type A cells give rise to intermediate spermatogonia which are present from late stage II to about stage IV when they again divide to form type B spermatogonia. Type B spermatogonia divide in stage VI to form resting primary spermatocytes. These are present during stage VII and during the first half of stage VIII; they then enter the leptotene stage of meiotic prophase.

Thus it is possible, by careful control of the timing of the exposure period, to irradiate specific cells in the spermatogenic cycle. In these experiments, the length of the exposure to the injected compound permitted effects on both intermediate and type B spermatogonia to be measured. These cells have been shown [19] to be amongst the most radiosensitive to cell death in the mouse. At the time of injection of tritiated thymidine, all the intermediate spermatogonia which were synthesizing DNA incorporated tritium into their nuclei. Subsequently when these cells had matured to resting primary spermatocytes in stage VIII the animals were killed and sections of the testes prepared for histological examination. The decrease in the numbers of resting primary spermatocytes per tubule, as compared with a control, then represented the radiation damage inflicted by the tritium. By reference to Roosen-Runge [27] based on work by Oakberg [20,21], the length of time from early/mid stage III (intermediate spermatogonia synthesizing DNA) to early stage VIII (resting primary spermatocytes) is about 90 hours (assuming no prolongation of the mitotic cycle due to radiation effects) [28]. Therefore, the length of the exposure to the various compounds used in these experiments was 90 hours, and during this time the cells under investigation had undergone two synthesis stages and two divisions.

Methods and Materials

DBA 2 male mice of 20 to 25 g weight were used throughout the experiments. These animals were fed MRC diet 41B and were allowed free access to water at all times. Twenty-five mice were used to provide control histological data.

All radioactive compounds were injected intraperitoneal in sterile isotonic saline. The injected radioactive compounds were all obtained from the Radiochemical Centre, (RCC) Amersham, England and were:

> Uridine-6T (3 Ci/mM) L-methionine-T(G) (150 mCi/mM), thymidine-6T(n), (37.8 Ci/mM and 5 Ci/mM), tritiated water, inulin-T (250 mCi/mM) and L-methionine ^{35}S (397 mCi/mM).

The injected solutions were calibrated against n-hexadecane 1,2-T (RCC) in the case of tritium and n-hexadecane-^{14}C in the case of ^{35}S.

In the experiments designed to measure biological effect, the animals were used in groups of three or four and injected with 2,5, 10 and 20 μCi/g body weight and killed at 90 hours as explained above. In one series of experiments, inactive thymidine at concentrations of 12.5 μg, 25 μg, 50 μg, 100 μg per gram body weight was used for comparison.

After sacrifice, at 90 hours following injection, both testes were removed immediately from the mice and dissected free of connective tissue. One from each animal was fixed in formol saline for 36 hours, and the other was assayed for tritium as described below. The organs were subsequently dehydrated with alcohol, cleared in chloroform and embedded in paraffin wax. Six sections were cut from each testis at a thickness of 5 μm separated by 50 μm to ensure that each contained a different combination of cells.

All sections were stained by the periodic acid-Schiff (PAS) technique using Lillie's modification of the Hotchkiss method [26]. Scoring of spermatocytes in stage VIII tubules was carried out at a magnification of 400 using only tubules cut perpendicularly to the basement membrane, i.e., whose shape approximated a circle, in order that cells at the same stage were present around the periphery of each tubule examined. All resting primary spermatocytes were scored if their nuclei were wholly or partly within the section but no obviously necrotic cells, e.g., cells with damaged cell or nuclear membranes were counted. In this way the resting primary spermatocytes in about 100 stage VIII tubules were counted per mouse.

In the experiments designed to measure the body turnover of the compounds, the animals were injected with 2 µCi/g body weight and killed in groups of four at daily intervals up to 20 days. In most experiments, the concentration of total tritium only was of interest as average organ dose was calculated. The testis was dissolved in 5 ml of 1M KOH at 60°C with constant stirring and, when dissolved, 5 ml of Hyamine-10X (1M solution in methanol) was added. One-tenth ml of this solution + 0.075 ml of 2N HCl was then mixed with 10 ml of phosphor (5 g diphenyloxazole + 100 g napthalene dissolved in 1 liter of dioxane) and counted for tritium or sulphur-35 as appropriate by liquid scintillation. Quench correction was by the external standard channels ration technique.

Results and Discussion

In most cases, it was found that the loss of activity from the testes could be conveniently represented by a single exponential function over the first seven to ten days following injection, shown in Figures 1 to 3. Inulin-T was anomalous in that uptake, even at one

Figure 1

Loss of Thymidine -^{3}H and Tritiated Water from Whole Testes of Mature Mice after Intraperitoneal Injection

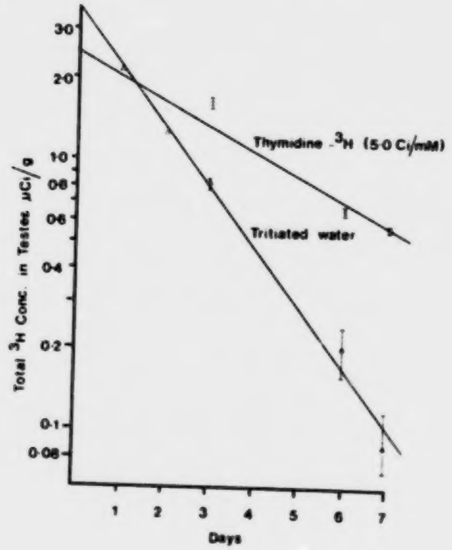


Figure 2

Loss of Methionine -^{3}H, Thymidine -^{3}H and Uridine -^{3}H from Whole Testes of Mature Mice after Intraperitoneal Injection

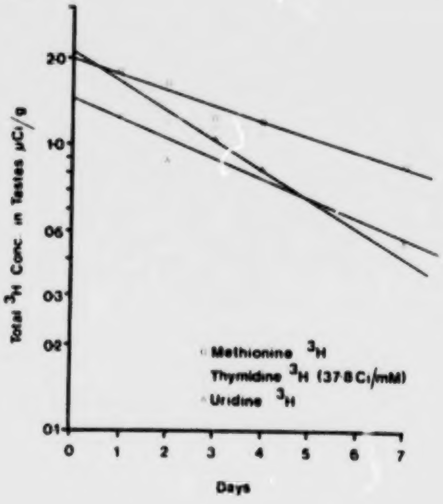
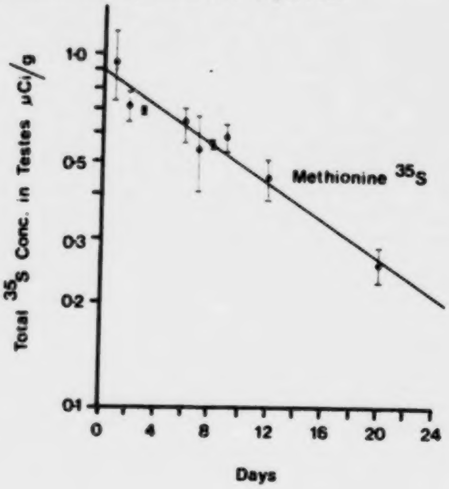


Figure 3

Loss of Methionine -35S from Whole Testes of Mature Mice after Intraperitoneal Injection



day, at all injection levels was only just significant and consequently no dose or biological effect measurements could be made. As was expected, the excretion of inulin was too fast to be detected by daily measurements. The half periods of loss of radioactivity from the testes are shown in Table 1 for the compounds studied. From this table it can be seen that there is no significant difference between the two specific activities of tritiated thymidine used. Other authors [29] have shown an almost constant level of (nonvolatile) tritium in the testes following injection of tritiated thymidine. Presumably the loss of tritium demonstrated here is the loss of tritiated water of catabolism due to turnover of the labeled compound in the liver, superimposed on an approximately constant amount of tritium incorporated into DNA. This concept is further exemplified by the half-lives of methionine-T and methionine-^{35}S. In the case of tritiated methionine, the tritium is fairly easily removed as tritiated water, whereas the sulphur-35 is more firmly bound (e.g., in protein) and this longer half-life more truly represents the turnover of methionine.

The data in Figures 1 to 3 were used to calculate the average doses to the testes using the formula:

$D = 51.2\ E.\ I$ rad [30]
where E is the effective energy of the β particles per disintegration of tritium (5.7×10^{-3} MeV) [31], and I is the time integral of activity per g (in μCi-days per g).

The average organ doses following an injection of 2 μCi of the various compounds/g body weight are shown in the table. These doses show a remarkable similarity (except that from ^{35}S-methionine) which could be a reflection on the lability of the tritium label. However, the damage inflicted on the cells of the testes, which is shown in Figure 4, suggests that average organ dose might be used to a first approximation to predict this damage. For example at 20 μCi injected/g body weight (which for a 20 g mouse implies 400 μCi injected), the depression in the numbers of resting primary spermatocytes/tubule varies by a factor of five among the compounds studied, except for uridine-T. Even for the most toxic compound studied, high specific activity tritiated thymidine, the numbers of resting primary spermatocytes scored 90 hours after injection of 20 μCi/g body weight was only about 50% when compared with the controls, in which 67.3 ± 9.7 resting primary spermatocytes/tubule were scored.

In fact, as the radiation doses reported here were so similar, it might at first be assumed that merely the effect of the tritiated water of metabolism was being measured by the experiments. This could be so if the uptake into the organic fraction of the cell was small. However, the half periods of elimination of the compounds injected were all significantly larger than tritiated water itself, see Table 1. In addition, in two cases the tritiated water of catabolism was measured and half periods of elimination determined. After injection of methionine-T the half period of elimination of volatile materials (tritiated water) was found to be 2.0 days, which is similar to tritiated water itself, and after injection of uridine-T was 4.1 days. This is similar to the elimination of total tritium in this case, i.e., there was maximum uptake into organic species. In the case of tritiated thymidine, previous work [18] involving the measurement of both volatile and nonvolatile tritium fractions in the testes has shown that the radiation doses from DNA-T and tritiated water averaged

Figure 4

μCi of Compound Injected per g. Body Weight

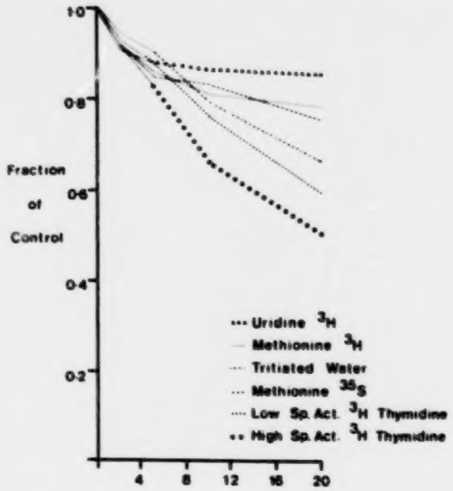


Table 1

Depression in the Number of Resting Primary Spermatocytes per Tubule for Mature Mouse Testes after Intraperitoneal Injection of Radioisotope Labeled Compounds.

Compound Injected	Half Period of Elimination From Testes (Days)	Radiation Dose (rad)*
Tritiated water	1.4 ± 0.2	1.8 ± 0.2
Thymidine-^{3}H (37.8 Ci/mM)	3.0 ± 0.4	1.5 ± 0.2
Thymidine-^{3}H (5.0 Ci/mM)	3.3 ± 0.4	1.9 ± 0.2
Methionine-^{3}H	5.5 ± 0.3	1.8 ± 0.2
Uridine-^{3}H	4.4 ± 0.4	1.2 ± 0.1
Methionine-^{35}S	11.7 ± 0.5	8.6 ± 0.4

*Average dose to the testes over 90 hours as a result of injection of 2 μCi of the various compounds/g body weight.

throughout the organ were in the ratio of about 1.5:1 and the radiation doses to the nucleus of a dividing intermediate type spermatogonia [14] from DNA-T and tritiated water were in the ratio of about 6.4:1. Thus, following injection of tritiated thymidine, it is apparent that there is a significantly larger radiation dose from nonvolatile tritium, irrespective of the volume over which this is averaged within the testes.

The experiment, as carried out, measures the effect on intermediate and type B spermatogonia, in particular their ability to divide twice and produce resting primary spermatocytes. The timing and design of the experiments suit the measurement of acute biological effect following the injection of material utilized, e.g., tritiated thymidine, during the synthesis stage of the cell cycle. Materials utilized throughout the cell cycle or at other points during the cycle will generally irradiate slightly different populations of cells. However, from the point of view of practical radiological protection, the concept of average organ dose is easiest to use, and it is apparent from the results that for tritium, at least within a factor of five, it could be utilized to estimate the acute hazard. Nevertheless there are two anomalies which require some explanation.

1. Uridine-T produced far less damage than the other compounds relative to injected activity. However, consideration of the error on each point in Figure 4 indicates that there is no significant difference between the curves for tritiated uridine, tritiated methionine and tritiated water. Also it can be seen from the table that uridine-T does deliver a significantly smaller average testes dose than the other compounds.

2. The dose from methionine-^{35}S is considerably greater than the other compounds studied and does not reflect the damage produced in the testes. This is surprising since, of all the compounds studied, methionine-^{35}S should have given the most homogeneous dose pattern throughout the organ due to its distribution and the higher energy of its β-emission. Thus, it would seem that the localization of tritium in certain sensitive volumes of the cell produces a large concentration of absorbed energy which results in local damage and subsequent death of the cell. This indicates, even though the results of experiments with tritium labeled compounds suggest the contrary, that biological effect should be related to the dose to some small sensitive volume of the cell, but the question should perhaps remain open until further investigations have been carried out using other ^{14}C and ^{35}S labeled compounds. One study has been carried out using both ^{14}C and T compounds in vitro [32], but no significant work has been reported in vivo.

In addition to the radiation effect experiments reported in Figure 4, the effect of "cold" thymidine was also tested using this system. However, even at 100 μg thymidine injected per g body weight, the depression in the numbers of resting primary spermatocytes was less than 10%. Bearing in mind that 20 μCi of tritiated thymidine (5 Ci/mM) injected per g body weight is equivalent to only about 100 μg of thymidine, a correction factor was not used to account for the thymidine "pool effect."

Conclusions

The main and most important conclusion which may be drawn from this work is that, to a first approximation, there is no great difference in cell-killing effects during 90 hours exposure between the compounds studied relative to injected activity. This conclusion has been reached before both in review [10] and based on work carried out here [14].

The results are also a further demonstration that the seminiferous epithelium is a most sensitive system when studying cell death. In the experiments reported here, all the compounds produced just significant cell death at an injected level of 2 μCi/g body weight (i.e., about 50 μCi total tritium injected). Considering the different body masses and difference in turnover rates, this would be equivalent to about 20 mCi of tritiated compound injected into man. However, it has been shown [10,18] that for tritiated thymidine, the maximum permissible annual intake is about 20 mCi and it seems unlikely that this level would ever be reached in practice. In addition, it is dangerous to make assumptions and draw conclusions from animal data which allow absolute estimates of the hazard to man to be made. Thus, the results of the experiments reported here should be used as a means of comparison with, say, tritiated water.

In general it is evident that the cell-killing effects in the testes show that, to first approximation, one could use average organ dose to predict damage from the tritium compounds used. More precise considerations would indicate that the spatial distribution of the tritium label, and in particular its location relative to radiosensitive sites within the cell, would chiefly control the damage. In practice it is difficult, if not impossible, to estimate dose to subcellular volumes and, therefore, sufficient in practical radiological protection to consider average organ dose. However, this work only applies to this particular cell system and particularly cell death within the system. The results may not apply to other effects, e.g., mutagenesis or carcinogenesis within this organ or other organs.

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TRANSMUTATION VERSUS BETA IRRADIATION IN THE PATHOLOGICAL EFFECTS OF TRITIUM DECAY

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There is hardly a problem in radiobiology more in need of clarification than the biological effects of transmutation of incorporated tritium [1]. Information adequate for detailed analysis is available only in relatively few experiments.

An earlier survey of data available on killing efficiency of incorporated tritium in mammalian cells led to the conclusions [2,3] that:

1. The origin of tritium beta tracks in, or their close juxtaposition to, the DNA molecule does not appear to enhance the degree of somatic effect.
2. The distribution of tritium atoms incorporated into DNA as tritiated thymidine (and of ion pairs from the beta-particles) is random as far as dose calculations for the purpose of predicting somatic effects are concerned.
3. Every part of the cell nucleus lies within one tritium beta range of some part of a chromatid, and the nucleus contains no sizable contiguous "insensitive" volume of a radius exceeding the effective range of tritium beta rays (1 to 2 μm).

Also, studies on mutation induction from incorporated tritium in *E. coli* and bacteriophage have revealed tritium transmutation effect only if the tritium decay occurred at the 5 position of cytosine in DNA [4]. Only about 2% of the DNA hydrogens and 0.04% of the total nuclear hydrogens are located at this specific position.

Since these observations and conclusions, new data have become available and possible mechanisms of transmutation effects have been better defined. It is the object of the present paper to see whether in the light of these new data the earlier conclusion should be modified.

Tritium Decay — Basis for Transmutation Effects

Tritium decays in a single step to helium and emits a beta-ray with a maximum energy of 18.6 keV and a maximum range of about 6 μm in water. The average energy is 5.7 keV, and the average range is less than 1 μm in water. Ten percent of the betas have energies of less than 1 keV, and approximately 1% have less than 100 eV and are absorbed over a range of about 20 Å. The energy range of the tritium beta-particles is comparable to the energies of Compton electrons produced by X-rays [3].

In addition to ionization and excitation along the beta tracks within a volume of less than 100 pg, there are events strictly localized to the site of the tritium decay. These are initiated by the chemical transmutation of tritium to helium and to some degree by the residual electronic excitation from the sudden acquisition of a positive charge of the daughter nuclide. The maximum nuclear recoil energy from tritium decay is approximately 3 eV. Since not all recoil energy that is calculated for a single nuclide can be expended in bond rupture [5], the maximum of 3 eV recoil energy imparted to the helium nucleus is too low for breaking a carbon-hydrogen or carbon-carbon bond [6].

It is a particular characteristic of tritium decay that it always leads to the separation of the daughter nuclide helium from its bonding partner carbon. This is due to the chemistry of the helium-carbon bond, the energy of which is only 0.05 eV [7]. The positively-charged helium, following decay of tritium remains bonded to carbon for less than 10^{-4} to 10^{-5} seconds. The helium takes an electron from the carbon and becomes electrically neutral, leaving the carbon positively charged. This may act as a free radical ion, the fate of which varies greatly according to its position in the molecule and with the chemistry immediately around it.

The inevitable breakage of the carbon-helium bond within about 10^{-4} to 10^{-5} seconds is preceded by electronic excitation of the newly formed helium. This amounts at average to approximately 11 eV [3]. Since the positive charge of the helium is produced within a relatively short time compared to orbital electron motion, the resulting excitation energy is expected to be partially transferred to the carbon atom prior to breakage of the helium-carbon bond. The extent of transfer of excitation energy to the positively charged carbonium ion is not clear. However, a particular effect on the bonded partner appears to be small, since in diatomic tritium molecules, 58% of the tritium decays were found to yield diatomic daughter ions without electronic excitation [8].

It appears justifiable to predict that biological effects of tritium transmutation are mainly a consequence of the production of helium, which inevitably results in the formation of a positively charged carbonium ion. The particular site of this carbonium ion appears important; numerically it adds to the approximately 180 ionizations caused by the tritium beta-particle of average energy. Because of the short range of the low energy beta-particles, transmutation effects of tritium are not easily recognized in complex molecular systems within biological structures.

Transmutation Effects and Cell Killing

Tritium bound within the DNA of bacteria was found to be more lethal than tritium positioned within the protein around the DNA core [9,10]. An example is shown in Figure 1. The killing efficiency in *E. coli* for tritium-methyl-thymidine in DNA was 2.9%; it was 40 to 50% of this value, namely 1.5 to 1.1%, for 5-tritium-uridine and for tritiated histidine. It is important that the killing efficiency of tritium located in position 5 of uracil was practically identical to that of tritium in position 6. It also made no difference in killing

Figure 1

Killing Efficiency in E. Coli for Tritium-Methyl-Thymidine [9]

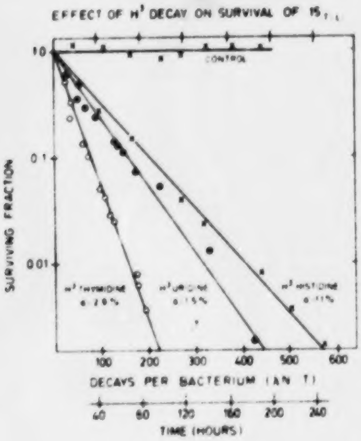


Table 1

Deposition of Energy (eV) in Central Core [11]

Labeled Region	
Small nucleus	1018
Large nucleus	924
Average nucleus	971
Whole cell	354
Cytoplasm	183
Middle cell	229
Thin cell wall	104
Thick cell wall	95
Average cell wall	100

efficiency whether thymidine carried tritium in the methyl group or in the 6th position [4]. The particular toxicity of tritium bound to the bacterial DNA was initially viewed as evidence for a transmutation effect. However, these results could be explained by a more efficient irradiation of the bacterial DNA localized in a central core. Depending on the location of the tritium in the various volumes around the central bacterial core, different amounts of energy are absorbed within the core, as shown in Table 1 [11,12]. It is clear that the central core, the site of the bacterial chromosome, absorbs about ten times more energy from the tritium beta originating within the core than from the beta track originating in the cell wall.

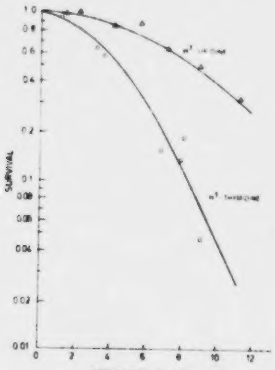
A similar correlation between site of tritium incorporation and rate of biological inactivation was found for T_4 bacteriophage and could be explained entirely by different geometries of the beta-particle tracks [13]. This indicates that viability loss in bacteria and phage can be entirely explained on the basis of the beta radiation without recognizable additional effect from the tritium transmutation to helium [14,15].

No such exact statements could be made from data obtained with mammalian cells. But the principal effect of the geometry of the beta-particle track was again observed. In Chinese hamster fibroblasts, tritium incorporated into DNA of the cell nucleus was about 30% more lethal than tritium incorporated into RNA throughout the cell. Figure 2 shows the multiple hit dose effect curves for these experiments [16] carried out under normal culture conditions. It is obvious again that the sensitive target in the cell is the nucleus.

Thus, with respect to cell killing, the effects of tritium decay are explained adequately on the basis of beta irradiation, and no additional effect due to transmutation has been demostrated at normal temperatures. Below are discussions of studies in which damage is allowed to accumulate in the frozen state.

Figure 2

Survival of Chinese Hamster Fibroblasts after Irradiation from Tritium Incorporated in DNA (H^3-Thymidine) and RNA (H^3-Uridine) (Reference 16)



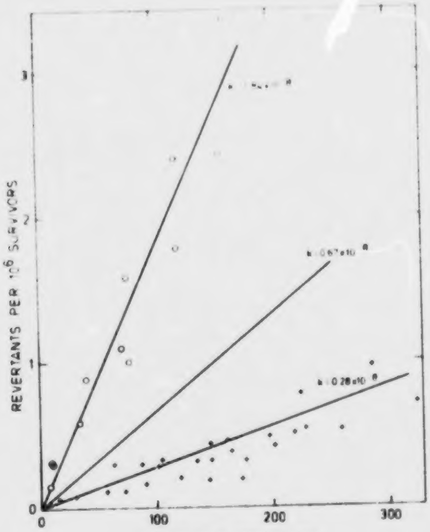
Transmutation Effects and Genetic Mutations

A first indication of a probable transmutation effect was reported by Rachmeler and Pardy [17]. These authors ascribed viability loss in E. coli exclusively to the beta-particle irrespective of whether the tritium was incorporated into RNA or DNA. The inactivation of the gene for betagalactosidase, however, appeared dependent on the location of tritium directly within that gene. Thus when the entire DNA was labeled, enzyme formation and viability were both affected. When, however, the DNA of the gene was spared from tritiation by mating labeled bacteria with unlabeled ones, there was no depression of enzyme formation.

Localized effects of tritium transmutation were unequivocally demonstrated when Person [18] examined mutation induction in E. coli following labeling with various tritiated precursors such as tritiated thymidine (tritium in the methyl group), 5-tritium-uracil, 6-tritium-uracil and tritiated amino acids. The mutation chosen in E. coli was the requirement for arginine. The bacteria were labeled for a period of several cell generation times, so that tritium decays finally occurred at a rate of approximately one to two per hour. The labeled cultures were then stored at 4 to 8°C in order to accumulate tritium decays. At various times during the storage period, samples were removed for testing survival, mutation, and rate of tritium decays.

Figure 3

Arginine Requirement Mutation in E. Coli after Cold State Irradiation by Incorporated Tritium Labeled Compounds [18]



The data are shown in Figure 3. The probability of one tritium decay to produce an arginine reversion (the so-called K-value) was about 0.67×10^{-8} for tritium-methyl-thymidine in DNA; for tritium bound to protein it was 0.28×10^{-8}. When 6-tritium-uridine was used for labeling the cells, the K-value was similar to that for tritium in protein. However, when the cells had incorporated 5-tritium-uridine, the probability of mutation induction rose nearly sevenfold to 1.84×10^{-8}. Despite the difference in mutation production between the two differently tritiated uracils, there was no significant difference in their lethal effects. Since the two labeled uracils showed no difference in incorporation rate or cellular distribution the large effect of 5-tritium-uracil on mutation induction could not be explained by beta irradiation.

Uracil is predominantly incorporated into RNA, and only 5 to 6% is converted in these bacteria to DNA cytosine [4]. Thus a specific mutation induction should be linked to effects on the DNA cytosine. It could be demonstrated that tritium decay in the 5 position of DNA cytosine in *E. coli* initiates a coding change from cytosine to thymine, whereas no such change was detected with tritium decaying in the 6th position [4].

These studies involved storage of labeled bacteria at low temperatures. In more highly developed systems useful for mutation analyses, such as *Drosophila*, low temperature treatment for the purpose of dose accumulation has not been carried out. When the cellular metabolism is not altered by low temperatures, repair mechanisms are expected to be effective and may perhaps lead to different results.

Data on mutation induction from tritiated nucleic acid precursors are not yet available in mammalian cells. Yet in *Drosophila* the mutation rate induced by uridine tritiated in the 5 position was twice as high as with uridine tritiated in the 6 position [19]. The data are shown in Table 2. Primary RNA alteration in the induction of recessive lethals could be practically excluded, since the effect was higher for labeled euchromatin than for heterochromatin, and since it occurred irrespective of involvement of nucleolar organizer regions.

Another study with tritiated uridines in *Drosophila* pertained not to mutation induction but to carcinogenesis [20]. Here the 6-tritium-uridine was twice as carcinogenic as 5-tritium-uridine. This may indicate and confirm the hypothesis that tumor induction and mutation induction are not necessarily linked events.

In both *Drosophila* studies referred to above, no measurements of the amount of incorporated tritium were made. Thus no quantitative dose-effect relation is established.

Table 2

Mutation Induction by Tritiated Uridine in *Drosophila*

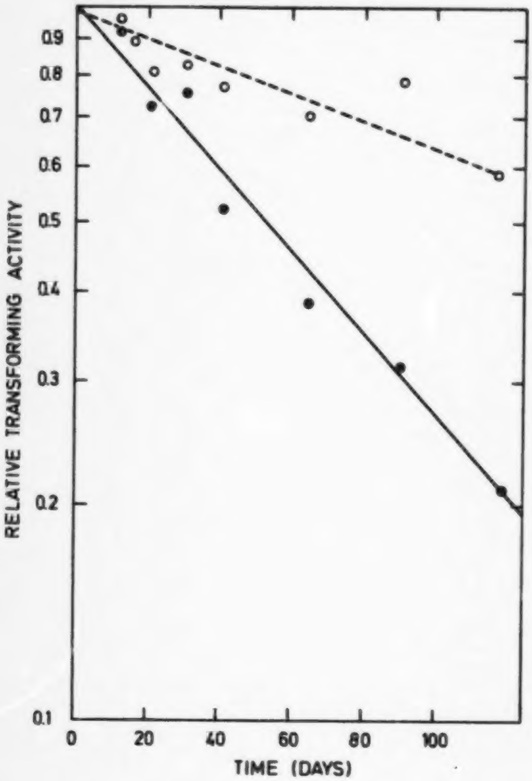
	5-T Ur. 1000 μCi/ml 5 Ci/mM						6-T Ur. 1000 μCi/ml 6.55 Ci/mM					
	Sex-linked leth.			2nd. chrom. leth.			Sex-linked leth.			2nd. chrom. leth.		
Brood	n	leth.	%	n	leth.	%	n	leth.	%	n	leth.	%
A	620	3	0.48	544	1	0.18	517	0	0	401	1	0.25
B	587	4	0.68	371	3	0.81	569	2	0.35	309	1	0.32
C	624	4	0.64	348	5	1.44	578	1	0.17	420	3	0.70
D	605	7	1.16	449	10	2.23	585	3	0.51	339	3	0.87
E	358	3	0.84	320	7	2.19	506	1	0.20	347	3	0.85
F	329	3	0.91	276	4	1.45	478	3	0.63	192	1	0.52
Total	3123	24	0.77	2308	30	1.30	3233	10	0.31	2008	12	0.70

Transmutation Effects in Single Strand DNA

Damage localized only to DNA may be tested directly using the transforming capacity of bacterial DNA. Thus, Rosenthal and Fox [21] used DNA from diplococcus pneumonia that was labeled with tritium in cytosine and thymine. This DNA preparation was placed in storage medium at -30°C for about 120 days to allow tritium decays to accumulate. At various times during storage, samples were thawed and examined for single- and double-strand breaks. Tritium decays produced single-strand breaks with an efficiency of 30% and double-strand breaks with an efficiency of less than 1%. The authors then made artificial hybrids of tritiated DNA resistant to streptomycin and

Figure 4

Streptomycine Transforming Activity of Tritiated and Non-Tritiated Bacterial DNA Stored at Low Temperatures [21]



nonlabeled DNA carrying another marker. In the course of storage of these hybrid preparations at low temperatures, the rate of loss of transforming activity of the DNA strand carrying tritium was found to be four to five times greater than that of the nontritiated strand. These data are shown in Figure 4.

From these results, it is clear that tritium in double-strand DNA stored at low temperatures probably inactivates by means of single-strand breaks. Single strands carrying tritium are broken four times more efficiently than are the complementary unlabeled strands approximately 20 Å away. This is strong evidence for transmutation effect, probably mediated through the formation of a radical carbonium ion produced in position 6 of the pyrimidine ring. Earlier studies with tritiated DNA labeled in one strand only were reported by McCarthy and Nester [22]. These authors labeled *bacillus subtilis* for one generation time with tritiated thymidine, tritiated histidine or tritiated leucine. The killing efficiencies for these various tritiated compounds were compared to inactivation of transforming DNA extracted from genetically marked cells following suitable storage at -88°C. The killing efficiencies for the various tritiated compounds were all 7.2 to 7.7 times greater than the efficiencies for gene inactivations. Thus if viability loss in this organism is totally ascribed to ionization and excitation from the beta particles, then gene inactivation in these experiments *in vivo* appears to be mediated by the same mechanisms.

Toxic effects of tritium decay in single-strand tritiated DNA were also observed in a mouse leukemia cell L 5178 Y stored at -196°C. Burki and Okada [23] compared the survival of cells labeled with tritium-methyl-thymidine either in one strand of DNA only (unifilar labeling) or at random in both single strands (bifilar labeling). After various storage times at -196°C, the labeled cells were rapidly thawed and placed in normal medium for viability test. The growth curves of the cells in regular medium were used to calculate survival as a function of tritium decays and of rads to the cell nucleus from the beta radiation. With unifilar DNA labeling, the dose effect curve appears exponential. The D_{37} value is lower than that obtained with cells that were labeled in both strands at random, and this again was lower than the D_{37} from RNA bound tritium or from cobalt-60 gamma irradiation. The experiments of Burki and Okada indicate that the killing efficiency of tritium in DNA at -196°C is more effective than it is under normal culture conditions. Moreover, it is obvious that effects localized to the site of tritium decay must have accumulated in the frozen state.

Discussion and Conclusion

Tritium transmutation effects have now been well established. As far as cell killing is concerned, the transmutation effects have been recognized at low temperatures when normal repair processes were inhibited. Under normal growth conditions, the calculated dose to the cell nucleus from DNA bound tritium for a given degree of effect is comparable to that from external irradiation for the same degree of effect. Thus, irrespective of the mechanisms involved that lead to cell death, tritium decays within the DNA molecule under normal growth conditions do not appear to cause observable effects in addition to those from the beta radiation. That means that transmutation effects from tritium in DNA appear to be repaired with a high degree of efficiency in the normal state.

A more quantitative evaluation is possible in the light of the data presented above relating to damage accumulation in the frozen state. Tritium transmutations in the 6 position of the pyrimidine ring in single-strand DNA causes single-strand breaks with an efficiency of about 30% and double-strand breaks with an efficiency of less than 1% [21].

Two to three tritium decays in a cell nucleus of 6 to 7 μm diameter, deliver to that nucleus a dose of 1 rad average, or approximately 350 to 400 ion pairs. These also cause single- and double-strand breaks irrespective of other effects such as base changes. According to Veatch and Okada's work with L 5178Y cells, the nuclear diameter of which is 6 to 7 μm, 1 rad produces three single-strand breaks and 0.3 double-strand breaks per cell [24].

It is clear then that two to three tritium decays in DNA per cell increases the number of single-strand breaks to approximately four, and double-strand breaks from approximately 0.3 to 0.33 per cell. Single-strand breaks are known to be rapidly and efficiently repaired under normal growth conditions. Biological effects from single-strand breaks can therefore be neglected here. There is also indication that double-strand breaks may undergo repair. It appears justifiable to conclude that biological effects to the genetic material from tritium decays within the DNA molecule are increased by not more than 10% of the damage from tritium decaying at random elsewhere in the cell nucleus. At low dose levels, allowing one tritium decay per cell nucleus, there will be approximately one to two single-strand breaks per nuclear DNA; every tenth cell with one tritium decay in the nuclear DNA will exhibit a double-strand break from the beta irradiation; and every hundredth cell under the same condition will show one double-strand break from transmutation. This additional damage is not easily recognized experimentally.

Damage from tritium transmutation observed in tritiated DNA at low temperatures is obviously not as efficiently repaired as under normal growth conditions. This suggests that the predominant transmutation effect from the tritium bound to carbon in the DNA is the production of a reactive carbonium ion that may be regarded in principle to be equivalent to ions produced by irradiation. Thus when about 180 ionizations are produced per tritium on the average, there is only an insignificant numerical addition from the process of tritium transmutation to helium. The site of the positively charged carbonium ion however is important. Tritium in the 6 position of the pyrimidine ring produced no increase in mutation, whereas tritium localized solely in the 5 position showed a significantly increased effect. Accidental substitution of hydrogen at this position with tritium, for example from injected tritium water, is expected to be rare, since this position binds only approximately 0.04% of the total nuclear hydrogen.

In the light of more recent data, the conclusions reached earlier do not need modification. The effects of intracellular tritium are overwhelmingly due to beta irradiation of the nucleus. Transmutation effects do not produce a measurably increased effect under most conditions, and are detectable, if at all, only under highly specialized laboratory conditions.

NOTE

The evaluation and conclusions reported in this paper are a subject of discussion and contribute to the work by Committee 24 of the National Council on Radiation Protection and Measurements.

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THE QUALITY FACTOR FOR TRITIUM RADIATION

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The International Commission on Radiological Protection (ICRP) originally recommended a value of 1.7 for the quality factor (Q) of electrons with E_{max} less than 30 keV [1]. This was based upon the LET and the experimental RBE values of tritium, the only important nuclide in this energy range. Later, in view of the uncertainties inherent in the RBE values and in the interest of simplicity, the Q was lowered to unity [2-5]. This paper reviews the experimental evidence for the RBE of the tritium beta-particle with emphasis on the uncertainties involved. Taking these uncertainties into account, it appears that the RBE in a number of mammalian systems is closer to 2 than to 1. Since the Q is used in calculations for radiation protection standards, it seems more sensible to tend toward the conservative side of uncertainty and to use a rounded-off value of 2 rather than 1 for the Q of tritium beta radiation.

LET$_\infty$ of Tritium Beta Radiation

The values of Q recommended by the ICRP are related to the LET$_\infty$ or stopping power as shown in Figure 1. The initial LET of an average ^{3}H beta-particle (E = 5.7 keV) is about 3.5 keV/μm in water. The LET increases thereafter as the electron loses energy, giving an average LET over the whole track which has been estimated to be about 5.5 keV/μm. This estimate does not consider any possible redundancy in the track, however. The effective track width will depend upon the sizes of biological targets. Each time an electron track doubles back upon itself and takes a second pass through a biological target, it increases the LET, i.e., the effective LET as far as biological effects are concerned. In the extreme case, the track will be entirely redundant and the effective track length will be equal to the range of the electron (Figure 2). The mean range of a 5.7 keV electron in water is about 0.68 μm. If the energy were distributed evenly over this distance, the LET would be about 8.25 keV/μm in water. Referring to Figure 1, a range of possible LET values between 5.5 and 8.25 gives a range of possible Q values from 1.6 to 2.3.

Figure 1

The Relationship Between LET and Quality Factor as Recommended by the ICRP. The QF for the Tritium Beta Particle Lies Within the Range Indicated.

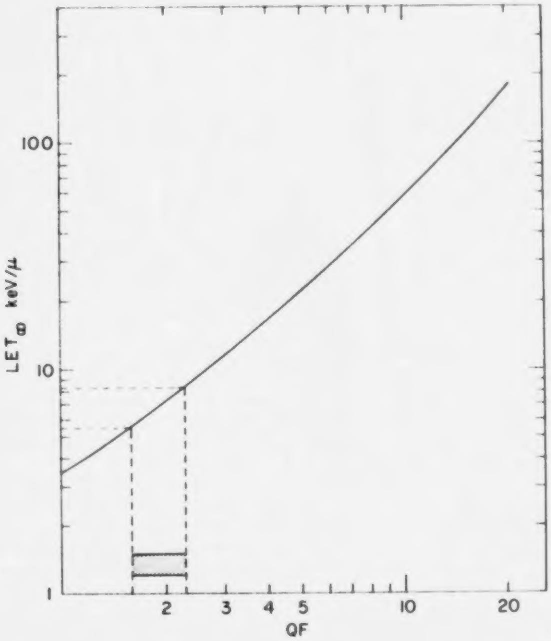
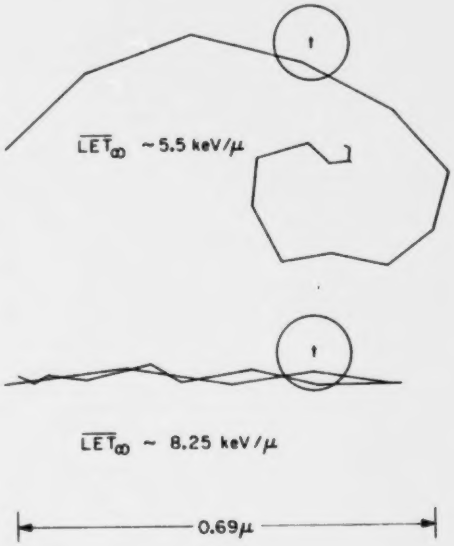


Figure 2

The Effective LET Depends Upon the Size of the Biological Target and the Redundancy of the Beta Track


$\overline{\mathrm{LET}}_{\infty} \sim 5.5$ keV/μ
$\overline{\mathrm{LET}}_{\infty} \sim 8.25$ keV/μ
0.69μ

Uncertainties in RBE Calculations

The experimental RBE is calculated as the ratio of doses of radiations of different qualities which are required to produce the same biological effect.

$$\text{RBE} = \frac{\text{Dose of reference radiation}}{\text{Dose of tritium radiation}}$$

It is to be expected that the value of RBE will vary somewhat from one biological endpoint to another, but what is far more troublesome is the fact that for any given endpoint there are many uncertainties inherent in the calculation of doses. Some of these are outlined below.

Reference Dose:
1) Gamma- vs. X-rays
2) Dose rate

Tritium Dose:
1) Mean energy
2) Effective half-life
3) Distribution of ^3H

Reference Dose

1) Most experimental values of the RBE for ^{60}Co gamma-rays relative to 250 kVp X-rays are close to 0.85. Thus the RBE of ^3H can be changed by a factor of 15% depending upon whether gamma- or X-rays are used for the reference radiation.

2) Dose rate. For proper determination of RBE, the dose rates of ^3H and reference radiation should be comparable. Ideally the reference radiation should be given at an exponentially decreasing dose rate corresponding to the turnover of tritiated water if the latter is used as the tritium carrier. Even if both tritium and reference radiation are given at the same rate, the experimental RBE can vary depending upon the dose rates of both radiations. Hall et al. [6] have shown a twofold increase in RBE of tritium relative to gamma-rays in growth suppression and killing of HeLa cells when the dose rate is decreased from about 32 rad/h to 1 rad/h. From this it appears that the beta radiation of tritium has less dose rate dependence than the lower LET gamma radiation.

Tritium Dose

1) Mean energy. The generally accepted E_{av} for tritium beta-particles is 5.7 keV, although values of 5.5 and 6.0 keV have been used for dosimetric calculations. This can cause up to a 5% variation in the calculated RBE.

2) Effective half-life. In experiments where the dose is accumulated from a single injection of tritiated water, the effective half-life of tritiated water is an important determinant. This is complicated by the fact that in toxicity studies, the water balance is altered by the radiation sickness of the animals, increasing the half time of ^3H with increasing dose [7]. Effective half-lives of ^3H administered to the mouse as tritiated water and measured by different techniques give values in the range of 1.75 to 2.3 days for the fast component [7-9].

3) Distribution of ^3H. Distribution factors are vague enough when ^3H is distributed in water, but they become far more complex for tritium bound to other metabolites. In the case of tritiated thymidine, for example, tritium is incorporated only into nuclei of cells in the S phase of the cell cycle, and then is diluted by cell

division at different rates in different tissues [2,10]. Because of their much greater simplicity, only estimates of RBE based upon tritiated water will be considered here.

The central problem in calculating the dose from tritiated water usually amounts to: given a certain average whole-body dose or given a certain level of radioactivity in body water, what is the dose absorbed by critical cells and their nuclei? Dose calculations are customarily based upon the water content of soft tissues, which is taken to be 0.70, 0.75, or 0.80. This in itself gives an additional arbitrary variation in the calculated RBE, but in principle it is not correct to use the tissue water content at all. Since the range of half of tritium beta particles is less than 0.68 μm [11], the dose to cells is due largely to that from intracellular water. The important factor, then, is not the concentration of water in tissues, but the concentration within the cells of interest or, perhaps more important, the concentration within their nuclei. The concentration of water within cells may be quite different from that in tissues. A considerable part of tissue water is interstitial so that one might expect the concentration of water in the cell to be less than that of the whole tissue. This would tend to lower the calculated dose to the cell and would tend to raise the calculated RBE above those based upon tissue water.

The water content of the cell is difficult to measure and probably differs from one cell type to another. By comparing the wet weight of isolated single neurons with the dry mass as measured by micro-x-radiography, Hydén [12] estimates that the neuron is about 75% water. Using a different technique and a different cell type, Harrison [13] finds a lesser water content. From desiccation and analysis of bulk tissue he estimates the liver cell of the rat to be only 60% water. It is not hard to imagine sources of error in either of these methods, so that these figures are only approximate. As for the distribution of water between nucleus and cytoplasm, x-radiography shows no significant difference in dry mass per volume in neurons which are simply fresh frozen and dried [12]. The data of Mauer and Primbsch [14], which show a lesser dry mass per volume in the nucleus than in cytoplasm, are based upon cells which have been thoroughly leached out by fixation and embedding of the tissue. In the calculations which follow, it seems realistic to assume that nucleus and cytoplasm contain somewhere between 60% and 75% water and that the radioactivity per mass everywhere within the cell is 60% to 75% that of body water. Remember, though, that these limits are based upon only two cell types. It is conceivable that the nucleus of the small lymphocyte, for example, which is densely stained and has only one-fourth the volume of the hepatocyte nucleus, might have a water content of less than 60%.

In experiments in which the radiation dose from tritium is estimated by measuring the radioactivity of body water, a significant error will result from not including the radiation dose from organically bound tritium. After administration of tritiated water, initially about 3% of the total body activity is tissue bound, and after 30 days 50% to 70% is bound [9].

A Review of RBE Experiments in Mammals

1) *Mammalian cells in vitro*. Hall *et al*. [6] compared tritiated water and gamma radiation with respect to the suppression of HeLa cell growth rate. The radiation was administered at low dose rates, 0.5 to 3 rad/h, comparable to the dose rates in *in vivo* toxicity studies. Cells were considered to be 80% water, and the dose to the cell was thus considered to be 80% of the dose to the medium. This experiment gave an RBE of about 2. If the water content of the cells is somewhere between 60% and 75%, the calculated RBE should have been in the range of 2.1 to 2.7. If X-rays had been used as a reference, the corresponding RBE range would have been 1.8 to 2.3. This gives an overall range of uncertainty of 1.8 to 2.7.

These investigators also showed that with high dose rates (32 rad/h) the RBE tends toward unity. Other *in vitro* studies of RBE at high dose rates, such as those of Dewey *et al*. [15] at 60 rad/h, also show this effect, giving RBE values between 1 and 1.5. These experiments will not be reviewed here because the dose rates are much greater than any encountered in *in vivo* experiments or in practical radiation protection calculations.

2) Brues *et al*. The first toxicity study on whole animals from which an RBE for tritiated water can be calculated was carried out by Brues *et al*. some twenty years ago [8]. They measured the $LD_{50/30}$ of tritiated water for CF_1 female mice and found it to be very nearly 1 mCi/g of body weight when given in a single injection. They also measured the water turnover in these animals, which gives a rate constant of 0.4 day^{-1} for the first 4 days ($T_{1/2}$ = 1.7 days) and 0.64 day^{-1} thereafter ($T_{1/2}$ = 1.1 days). The accumulated dose can be calculated from

$$D_t = \frac{I}{k}\,(1-e^{-kt})$$

where D_t is the dose accumulated at time t, I is the dose rate at t_0, and k is the turnover constant. The initial dose rate is: (51.2) (E_{av} in MeV) (Concentration in μCi/g body weight) = 292 rads/day. For the first four days after the injection of 1 mCi/g,

$$D_4 \text{ days} = \frac{292}{0.4}\,(1 - 0.202) = 585 \text{ rads}$$

$$\text{After 4 days } D_t = \frac{(292)\ (0.202)}{0.64} = \underline{\ 93 \text{ rads}}$$

$$\text{Total absorbed dose} = 678 \text{ rads}$$

This is the average whole-body dose from a single injection of 1 mCi/g of body weight of tritiated water. If we assume, as the authors did, that their mice were 75% water, then cells which were 75% water received the same dose, or 678 rads. For cells with 60% water the total dose would be 543 rads. At that time Brues had no reference value for the $LD_{50/30}$ of X- or gamma-radiation delivered at an exponentially declining rate. Furchner [7] gives 1350 rads as the $LD_{50/30}$ for ^{60}Co radiation given at an exponentially decreasing rate. This gives an RBE based on gamma-radiation of somewhere between 2 and 2.5. The RBE relative to 250 kVp X-rays would be between 1.7 and 2.1. The overall range of uncertainty is 1.7 to 2.5.

3) Storer et al. In the experiments of Storer et al. [16], nearly constant levels of tritiated water were maintained in body water for five days. Three end points were used: atrophy of mouse spleen and thymus and depression of ^{59}Fe uptake in red cells of rats. Initial body water concentrations were established by intraperitoneal injection of tritiated water. Urine samples, considered representative of body water, were monitored daily. The dose to soft tissues was calculated by assuming these tissues to be 75% water, so that the activity measured in body water was multiplied by 75% to obtain the activity in the tissues. (The authors give as a source of error the fact that body water is not distributed homogeneously throughout the body. In fact this does not enter at all into their calculations of dose.) They use $E_{av} = 6$ keV, which tends to raise the calculated dose. Also, their measurements of radioactivity in body water do not record some 5% of organically-bound tritium.

In the mouse experiments they calculated the RBE of tritium in terms of radium gamma-radiation to be:

Splenic atrophy: RBE = 1.32
Thymic atrophy : RBE = 1.52

The 5% corrections for average energy and organically-bound tritium more oreless cancel out. Allowing for the uncertainty in intracellular water (60-75%) gives

Splenic atrophy: RBE = 1.32 - 1.65
Thymic atrophy: RBE = 1.52 - 1.90

The RBE of 250 kVp X-rays and ^{60}Co gamma-rays relative to radium gamma-rays was unity in both of these systems, so that RBE values of tritium relative to X-rays and ^{60}Co gamma-rays are as given above.

For depression of ^{59}Fe uptake by red cells in rats, Storer et al. obtained an RBE of 1.64 relative to ^{60}Co gamma-rays. Allowing for the uncertainty in cell water, the RBE becomes 1.64 to 2.05. The effectiveness of ^{60}Co gamma radiation relative to X-rays was not measured in this system. Assuming this to be 0.85 gives a range of RBE = 1.39. The overall range of uncertainty is 1.39 to 2.05.

4) Furchner [7] measured the RBE of tritium beta radiation using the 30-day mortality of CF_1 mice after a single intraperitoneal injection of tritiated water. The reference radiation was ^{60}Co radiation given at an exponentially decreasing rate. The effective half-life of tritium in animals receiving an $LD_{50/30}$ dose was about 50 hours. Here the E_{av} was taken to be 6 keV. Also the dose rates were calculated from the activity of body water, blood in this case, and do not take into account organically-bound tritium. This leads to an underestimation of dose which may be compensated by the overestimation due to the use of 6 keV as the mean energy. The whole-body doses for 50% mortality were 804 rads from tritium and 1350 rads from gamma radiation. The 804 rads represent a whole-body dose. The local dose depends again upon the local water concentration as compared with the whole-body water content. If the whole body is 75% water, then cells containing 75% water will also absorb 804 rads. Cells containing only 60% water, following Harrison's estimate of rat liver cells, received only 640 rads. This gives an RBE range, depending upon intracellular water, of 1.7 to 2.1 relative to gamma rays and 1.4 to 1.8 relative to X-rays. The range of uncertainty is 1.4 to 2.1.

5) Lambert [17] has compared the effectiveness of tritium

and X-rays with regard to the killing or injuring of spermatogonia in the mouse testis. This method, as developed by Oakberg [18], is a sensitive and accurate endpoint for measuring radiation injury at very low doses. Lambert found that 72 hours after the intraperitoneal injection of 20 μCi/g of body weight of tritiated water, the population of resting primary spermatocytes was reduced by 27%, equivalent to the effect of 30 rads of 200 kVp X-rays. The latter was given at an exponentially decreasing dose rate over a 72-hour period. The tissue activity of the testis was measured directly and the integrated dose was calculated. This method accounted for bound tritium as well as that in water. It also had the advantage of not requiring any assumption about the amount of water in the tissue. The radiation dose to the tissue from tritium was calculated to be 12.2 rads, giving an RBE of 30/12.2 = 2.4. We are still not certain that the water content of cells and nuclei was equal to the water content of whole tissue, so that the tissue dose may not represent the cell dose.

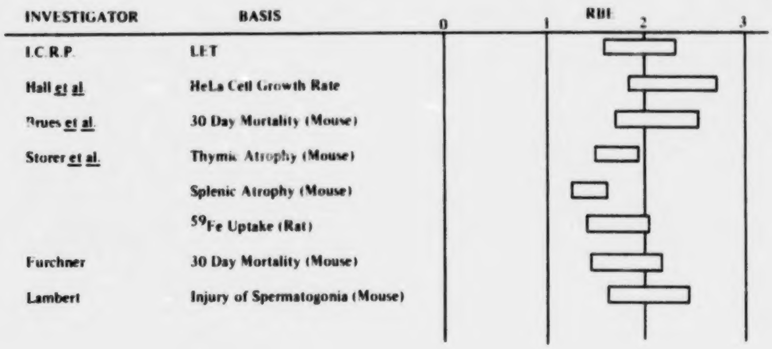
A similar experiment was carried out using tritiated thymidine. Here the dosimetry was based on autoradiographic grain counting. This probably gives a less accurate evaluation of the nuclear dose than does the dosimetry of tritiated water. This experiment gave an RBE of 1.6.

A Quality Factor For Tritium

The ranges of uncertainty for the various experimental RBE values are summarized in Figure 3. These values, however poorly defined, indicate a rounded-off 0 of 2 rather than 1. Add to this the fact that a quality factor which will be used in setting limits for radiation safety should take the conservative side of uncertainty, and it seems all the more appropriate that a Q of 2 be used in estimating the biological risks from a given absorbed dose of tritium beta radiation.

Figure 3

The Ranges of Uncertainty in RBE Data of Various Investigators



ACKNOWLEDGMENTS

Work contributing to the production of this report was supported in part by the U. S. Atomic Energy Commission.

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RELATIVE BIOLOGICAL EFFECTIVENESS OF TRITIUM

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Tritium is a soft beta-emitter, and a knowledge of its relative biological effectiveness (RBE) is of great practical importance. The available literature contains little information on the RBE of tritium, and that which is found is often contradictory [1-4].

The purpose of this work was to compare the effects of beta-radiation from tritium with the gamma-radiation from cesium-137 and thus determine the RBE. Experiments were performed using 587 rats. Tritiated water was given to animals in quantities of 3.0, 2.0, 1.0, 0.6, 0.3, 0.15 mCi/g body weight. Other rats were continuously exposed to gamma-radiation of cesium-137 in a special installation at the same incident dose rate as was calculated for the tritium [5].

The RBE of tritium was measured in terms of the following indices: survival rate, peripheral blood response, and thymic and splenic weight responses. Figure 1 shows the median survival rate as a function of tritium activity and of cumulative gamma-radiation dose. Massive doses of both tritium oxide and external gamma-radiation resulted in death of most of the animals within the first week of the experiment. Tritium appeared to be more effective than gamma-radiation. The dose causing death of one-half of the rats within the first four days was 1.86 and 3.72 kilorads for tritium and gamma-radiation, respectively. Hence, the RBE of tritium oxide is equal to two.

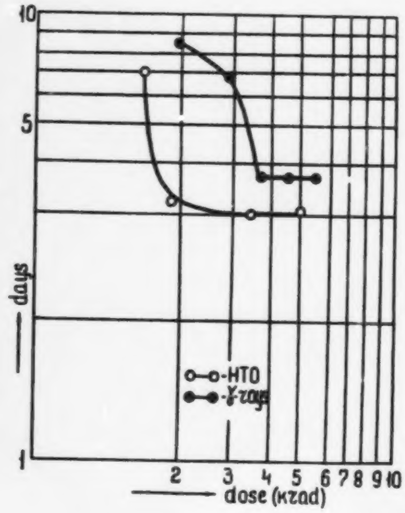
Injection of tritium oxide in a dose of 2 mCi/g body weight resulted in the death of 60% of the animals by the 9th day, when the tissue dose was 2 kilorads, compared with only 20% in the case of gamma-radiation. On the 12th day, after accumulation of a tissue dose of 2.2 kilorads, the mortality rate was 100% and 70%, respectively as shown in Figure 2. Following exposure to tritium oxide in a dose of 1.5 mCi/g and to an equivalent dose of gamma-radiation, 87% and 50% of the animals, respectively, succumbed by day 12 (the time of termination of irradiation) when the cumulative dose was 1.7 kilorads as shown in Figure 3. Differences in mortality rates persisted at later periods, using smaller doses (Figure 4).

On the basis of mortality rates with various doses of tritium oxide and external gamma-radiation, doses were calculated which caused death of 50% of rats by days 12 and 30. In the case of gamma-radiation of cesium-137, the $LD_{50/12}$ was 1820 ± 134 rads, and in the case of tritium 1350 ± 80 rads ($P = 0.05$). The $LD_{50/30}$ was equal to 1200 ± 43 and 895 ± 53 rads, respectively. These results show that the RBE of tritium is equal to 1.35 and 1.34. Thus, the RBE of tritium oxide lies between 1.34 and 2.0 using the survival criterion.

When used in acutely effective doses (3.0 to 0.6 mCi/g body weight or 3000 to 690 rads) tritium and gamma-radiation led to strongly

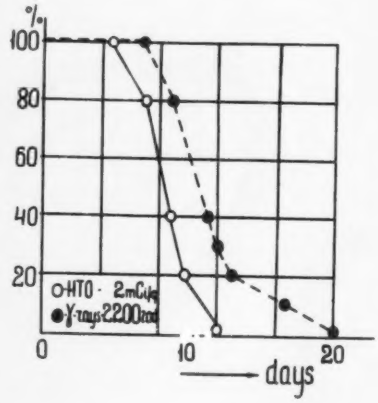
Figure 1

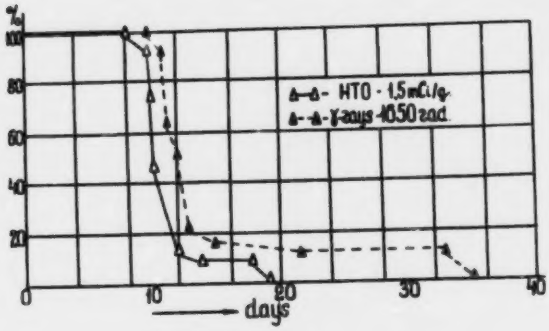
Rat Survival Time (Median) with Exposure to Gamma-Radiation of Cesium-137 or Tritium Used in Various Doses

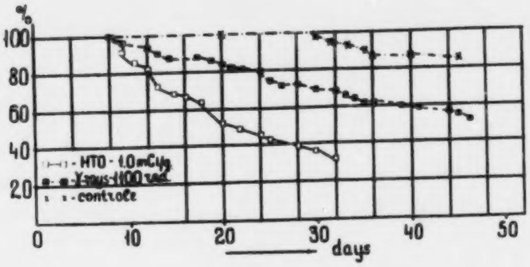


Figures 2-4

Time Course of Rat Death Rate Under the Effect of Gamma-Radiation or Tritium

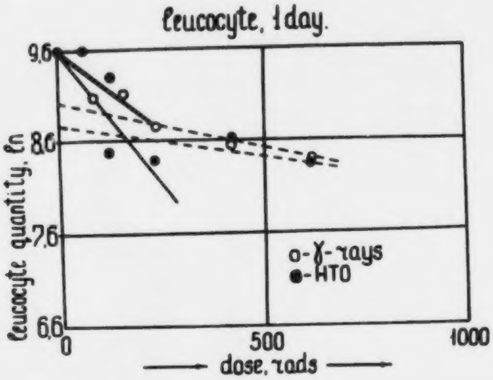


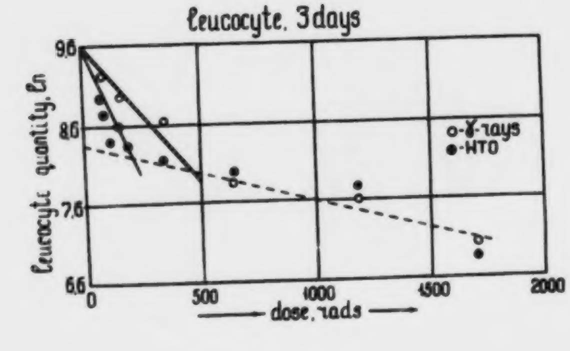




Figures 5-6

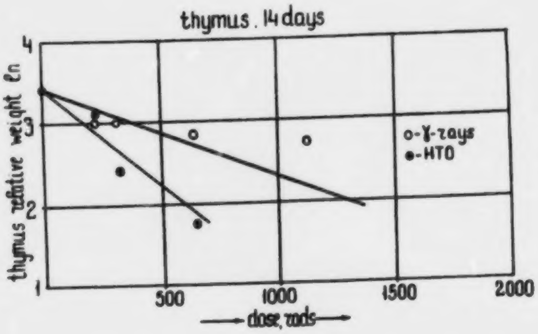
Dose-Effect Curves for Leukocytes on Days 1 and 3 After Exposure of Rats to Gamma-Radiation of Cesium-137 or to Tritium

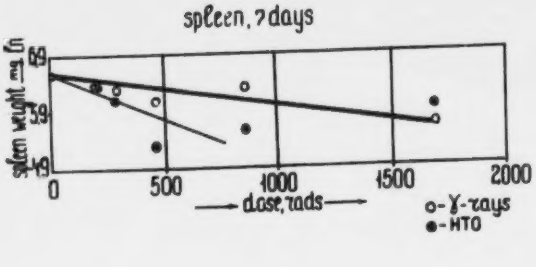




Figures 7-8

Thymic Weight Response on Day 14 and Splenic Weight Response on Day 7 as a Function of Dose Using Tritium or Gamma-Radiation of Cesium-137





marked leukopenia in the rats. The degree of leukopenia following exposure to equal doses was greater with tritium than with gamma-radiation. These differences were particularly great on day one. The dose causing a 50% decrease in leucocyte count was 224 rads for tritium and 426 rads for gamma-radiation. The dose causing a twofold decrease in lymphocyte count, one day after exposure, was 270 rads for tritium and 390 for gamma-radiation. The RBE of tritium is thus 1.45 (Figure 5).

With doses causing a decrease in lymphocyte count from 8000 to 3550-3000 per µl, the RBE of tritium oxide was between 1.67 and 1.90. In terms of thymic weight response, the RBE of tritium was 1.93 ± 0.37, at $P = 0.05$ (Figure 7). In terms of splenic weight response, the RBE of tritium on days three and seven was 1.74 and 1.89, respectively (Figure 8).

Analysis of the present finding has shown that, judging from the responses of peripheral blood and thymic and splenic weights, the RBE of tritium oxide lies between 1.45 and 1.93. Thus, in the case of acutely effective doses, tritium beta-radiation is biologically more effective than the gamma-radiation of cesium-137. The relatively greater biological effectiveness of tritium appears to be due to the fact that it develops 10 to 30 times as great ionization density per unit tissue volume as X- or gamma-radiation.

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TRITIUM OXIDE TRANSFER THROUGH PLACENTA, ITS INTAKE WITH MILK AND BIOLOGICAL ACTION OF TRITIATED WATER ON THE FETUS

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The transfer of radioactive isotopes from mother to fetus has grown into a problem which demands specific study. There are only a few facts in literature about the transfer of nuclides from mother to fetus through the placenta and into milk [1,2]. In this report, experimental data will be presented which indicate the level of placental transfer from mother to fetus and the level of transfer into the milk. Biological action upon the fetus will also be evaluated.

To study tritium oxide transfer through the placenta, the isotope was administered by ingestion (0.3 mCi/g body weight) to rats on the first day of pregnancy. The weight of the rats was 180 to 190 g. The kinetics of tritium exchange in mother and fetus were judged by measuring the concentration in tissues and organs on the 14th, 17th, and 22nd days of pregnancy, and by measuring the excretion of the isotope in the urine of pregnant rats. The method of treatment of the urine, tissues and organs was described earlier [3].

Results of the experiment showed that during different periods of pregnancy the tritium concentrations in the body water of the mother and the body water of the fetus were equal. These data indicate that the placenta does not act as a barrier for the tritium oxide transfer. The kinetics of the tritium oxide removal from the bodies of pregnant rats are described by the sum of two exponentials: 84 percent of tritium was eliminated during the first 2.7 days, and 16 percent during the next 0.1 to 0.2 days. Tritium concentration in the dry tissues of the fetus on the 22nd day of pregnancy was two to three times lower than that in the mother's soft tissues, Table 1. Fetal tissue concentrations were approximately the same as that in the mother's skeleton. Maximum tritium content in the fetus was at the 22nd day of pregnancy. At this time, the tritium content per fetus was 0.015 percent (0.15 percent in all offspring) of that ingested by the mother.

The calculated absorbed dose to the maternal body for 22 days of pregnancy was 1.02 rad, and to the fetus was 1.37 rad. The difference in the doses may be explained by the significant difference in body water content between the fetus and the adult (88 percent in the fetus to the 22nd day of prenatal development and 67 percent in the adult).

To study the kinetics of tritium exchange in the newborn, tritium oxide was administered (0.2 mCi/g) to pregnant rats before delivery. In this case, the concentration of tritium in the newborn of each litter was equal. The tritium concentration in the body water of the newborn (average from five litters) was 142±11 µCi/g. To the dry tissues, the tissue concentration was 6.8±0.4 µCi/g.

Experimental newborns were raised on milk from control female rats. On days 1, 3, 8, 15, 22, and 30 after birth, one offspring from

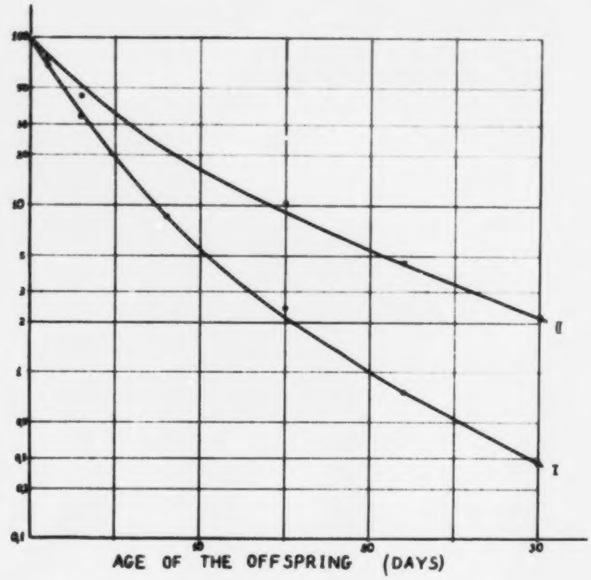
Table 1

Tritium Concentration in Dry Tissues of Rats on the 22nd Day After Tritiated Water Administration (10 μCi/g)

Rat	Organs and Tissues							
	Muscle	Liver	Blood	Skin	Bone	Uterus	Placenta	Fetus
Pregnant	3.4	2.14	2.2	3.0	1.3	2.6	1.26	1.18
Nonpregnant controls	3.1	1.9	1.88	2.8	1.18	-	-	-

Figure 1

HTO Concentration (I) and Content (II) of the Offspring in Percent HTO Concentration and Content at the Time of Birth



each female was sacrificed. In order that the number of feti nursing each female was constant (six) another offspring from another female was used to replace the one sacrificed. Figure 1 shows the change in tritium concentration and tritium content in the body of the offspring at different times after birth. Decrease in the tritium concentration takes place with two different half-lives, 87.5 percent with $T_b = 1.8$ days and 12.5 percent with $T_b = 5.6$ days, respectively. The change of the tritium content in the body of each offspring (Curve II) may be presented as the sum of two exponentials: 68 percent with $T_b = 2.2$ days and 32 percent with $T_b = 7.7$ days. By examination of the presented data, attention is drawn to the fact that change in tritium content in the newborn is slower than change in tritium concentration. This finding indicates that tritium in the offspring is diluted, because of rapid increase of the offspring mass.

In order to decide what has changed the kinetics of tritium removal from the offspring, a special experiment was conducted during the first 30 days of neonatal life. The animals remaining for this experiment were 20 females with six offspring each. The 20 females were divided into four groups of five each. During the suckling period, tritiated water was given to the offspring in the amount of 5.10 μCi/g (1, 7, 14, and 22 days after birth). At a different period after the tritiated water administration [one hour (zero point), one, three, five, and seven days] one offspring was sacrificed from every litter and the tritium concentration in body water was measured. The experimental data were used for defining the speed of the concentration change.

Results of this experiment showed that the tritiated water administered to the offspring on the first day is eliminated from the body with a half-life of 2.5 days, that administered at one week is eliminated from the body with a half-life of 2.3 days, and that administered at 14 and 22 days of the suckling period is eliminated with a half-life of 2.4 and 2.2 days, respectively. Taking into consideration that the exactness of the measurements could contain an error of 12 to 15 percent, the kinetics of the tritium elimination by the offspring for the first month appear to be practically equal.

To study tritium secretion in milk, tritium oxide was administered in the amount of 0.3 mCi/g to 25 adult rats on the first days of nursing. Each litter had six offspring. To determine the tritium content in the body water and dry tissue of the offspring at 1, 3, 8, 15, 22, and 30 days, five offspring from different litters were sacrificed. The sacrificed offspring from each of the lactating females, were replaced with other offspring from control groups. To study the tritium kinetics from the lactating nurse rat at different times, samples of blood were taken (about 0.2 ml) and studied to determine the tritium distribution at different times after isotope injection. The tritium content in tissues (muscles, liver, bone, and milk gland) was measured. The results of those experiments are shown in Table 2 and Figures 2 and 3.

The change of the tritium concentration in the body water of the nurse female (Curve I, Figure 2) occurred as an exponential with two T_b: 82.5 percent with $T_b = 2.9$ days and 17.5 percent with $T_b = 0.2$ days. In the offspring (Curve II, Figure 2), the tritium concentration in the body water is the same as that in the mother three days after injection. For the remainder of the 30 days, the tritium concentration in the body of the offspring is 1.5 to 2.0 times as high as that in the maternal body.

Table 2

Concentration and Tritium Content in the Maternal Organism, Offspring, and Whole Litter at Different Times in the Suckling Period

Tritium	Time after birth (days)					
	1	3	8	15	22	30
Tritium concentration in the offspring in percent of initial tritium concentration in the maternal organism	22.8	42.2	23.2	6.9	2.25	0.43
Tritium concentration in the maternal organism	65.6	42.1	13.5	2.93	0.72	0.24
The tritium content in the offspring in percent of tritiated water administered to the rat	0.394	1.26	1.21	0.495	0.191	0.055
Tritium content in the litter	2.36	7.56	7.26	2.97	1.15	0.33
Tritium content in the maternal organism	65.5	41.0	12.5	2.4	0.58	0.188

Figure 2

Specific HTO Activity in the Body Water of the Rats (1) and of Their Offspring (2) in Percent of HTO Injected Per Gram of the Body Weight of the Rats

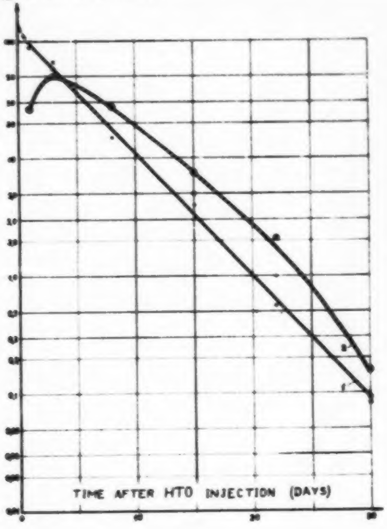
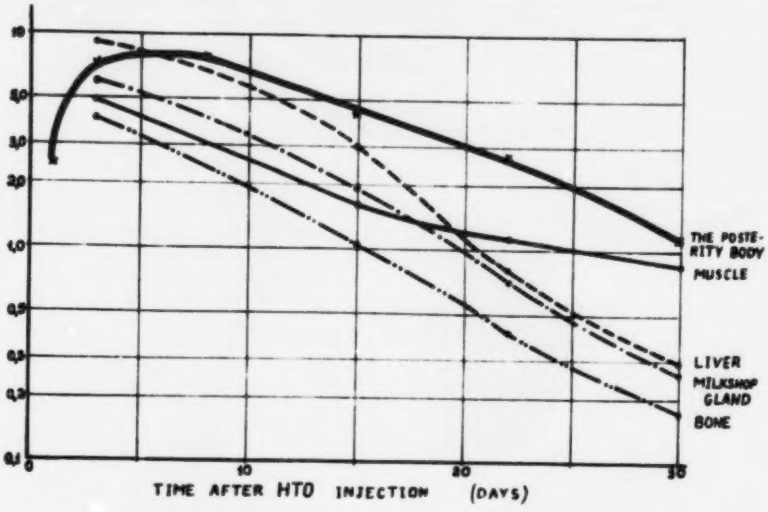


Figure 3

HTO Concentration in Dry Tissues of the Rats and of the Bodies of Their Offspring in Percent HTO Injected Per Gram of Bodv Weight of the Rats



Tritium concentration in the tissues of the offspring, Figure 3, matches the tritium concentration in the dry remainder of the maternal liver in five davs and, further, the tritium concentration in the offspring is higher than in the organs of the nurse females. Table 2 shows the tritium concentration in body water and dry tissue, and tritium content in the maternal organism, and of the offspring, at different times during the suckling period. Distribution of tritium between the body water and the dry remainder of the tissues in the offspring and in the mother were approximately identical.

In examining the data presented in this table, notice that the tritium content in the offspring at 30 days is three times lower than that in the nurse females. However, for the six offspring, the total content was 1.7 times as high as in the nurse females. It is evident that a significant amount of the tritium administered to the mother is transferred to the offspring in milk. Calculations showed, assuming constant milk volume intake by the offspring, that 2.2 percent of the tritium administered to the mother is transferred in the milk to one offspring. For litters having six offspring the total amount transferred is about 13 percent.

When tritium oxide was administered to rats (1 mCi/g) on the first day of nursing, the absorbed dose to the nurse female was 1.08 rad. The minimal absorbed dose accumulated in bones was 0.59 rad and maximal dose in the liver was 1.17 rads. The absorbed dose accumulated by each offspring for the entire suckling period is practicallv equal to the dose accumulated by the mother. However, the dry tissue dose to offspring was 25 percent greater than in the maternal body. The dry tissue dose makes up about 8.6 percent of the total absorbed dose.

Dynamics of the increase in the absorbed dose to offspring differ from the increase in the maternal organism. For first day in the maternal organism, 20 percent of the total absorbed dose is accumulated while in offspring only three percent is accumulated. During the first three days, the mothers received about 50 percent of the total absorbed dose and offspring about 20 percent. Two-thirds of the total absorbed dose was received by the mothers in five days; it took nine days for offspring to obtain this fraction of their total dose.

Results of the experiment showed that after single tritiated water administration in the amount of 0 3 mCi/g on first day of pregnancy, disturbances in the development of offspring were seen in both prenatal and postnatal periods of life. The observed disturbances in the embryonal period were embryo lethality and decreased fertility to 2.5 times that observed in the controls. Microscopic examination of the placentas and feti showed that tritiated water administration did not result in malformation. However, size and weight of offspring in the prenatal and postnatal periods were 1.5 to 2.0 times smaller than those in the control groups. Individual cases showed differences as great as three times smaller.

In the process of examining the feti and placentas, disturbances in the vascular system were found. These disturbances were vascular disorders in the placenta, and edema and subdermal hematomas in the feti. Also, duration of pregnancies was increased to 25 to 26 days instead of the normal 22 to 23 days in the control rats.

The pregnancy of experimental rats is often terminated at one of the two periods (25 to 26 days or 22 to 23 days). Of 28 pregnant rats (92 percent of the total bred) nine aborted, resulting in lethality of all the feti in the nine litters. The remaining 19 pregnant rats delivered 112 neonates, of which 20 were stillborn. Experimental offspring showed disturbances in normal development. During the postnatal period there was some delay (three to six days) in the development physiology of the progeny. The more prominent examples were delays in eye opening, teeth eruptions and hair covering. By 30 to 40 days, the progeny showed conjunctivitis, delayed development, and a decrease in weight of 20 to 40 percent. These conditions resulted in mass mortality (60 percent). A maximum number of lethalities were observed in the offspring which nursed for 30 days. The mortality seen in the control groups was only 50 to 60 percent of that exhibited by experimental offspring.

Thus, it was shown that the administration of tritium oxide in the amount of 0.3 mCi/g on the first day of pregnancy (absorbed dose during the period of pregnancy was 323 rads) resulted in pathology during prenatal and postnatal development. However, results indicated that this absorbed dose did not cause absolute sterilization effects, as only nine of the pregnant rats aborted resulting in lethality of the entire litter. It should be noted that tritium oxide administration on the first day of pregnancy resulted in the most lethality in the feti. Tritium oxide administered between 4 and 17 days of pregnancy did not produce as many fetal deaths (Lyaginskaya, 1966).

When tritium oxide (0.3 mCi/g) was administered on the first day of suckling, the offspring did not exhibit any changes in development as compared with the control groups. However, experimental offspring gained weight more slowly than control offspring, Table 3. On the 15th and 30th days, experimental offspring weighed 30 percent less

Table 3

Effect of the Tritium Oxide on the Body Weight and Mortality of the Offspring (Tritiated Water Administered in the Amount of 0.3 mCi/g on the First Day of Suckling)

The condition of the experiment	Numbers of litters	Number of the offspring in the litter	Weight of the posterity (g)			Mortality of the posterity at the suckling period	
			newborn	15 days	to 30 days	Abs. number	%
Experiment	10	8.2±1.1	5.9±0.87	18.9±0.9	35.2±5.2	10/82	12.2±1.2
Control	10	8.6±0.7	5.3±0.8	22.4±1.4	50.0±8.1	4/36	4.6±0.

than the controls. An increased lethality was observed also. During the 30-day suckling period, 12.2±1.2% of the experimental offspring died. In contrast, there were 4.6±0.3% lethalities in the control groups. Since the female rats and their offspring received approximately equal absorbed doses for 30 days (334 rads in offspring and 325 rads in female), these data can be taken as indicating that growing offspring are more radiosensitive than adult rats.

Results of this experiment showed that the histological barrier, the milk gland, does not prevent tritium oxide transfer from blood in milk. Tritium oxide was rapidly and easily transferred from the maternal organism to the nursing offspring via the milk. After the administration of tritium oxide (0.9 mCi/g) to the females, the increased absorbed dose in offspring resulted in decreased weight and increased lethality as compared to the control progeny.

It is apparent that there is a necessity for further experiments to study tritium oxide transfer from mother to fetus through the placenta as well as the transfer to milk. Also, the biological actions of tritium oxide on the growing organism require further study to verify the indications found in these experiments.

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TUMOR INCIDENCE IN C57 BLACK/6 MICE
TREATED WITH TRITIATED THYMIDINE

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The toxicity of tritiated thymidine (^{3}HTdR) and associated problems have been recently reviewed by Feinendegan [1]. This DNA precursor is of considerable interest because of its unique distribution and dosimetry. It is incorporated almost exclusively into the nucleus of cells synthesizing DNA prior to mitosis [2]. About 10% or more [3,4] of the injected ^{3}HTdR goes into the proliferating cell compartment, and remains incorporated into DNA until the death of the cell or its descendants. The remainder is catabolized to tritiated water and nonvolatile compounds [5].

The cell nucleus is selectively irradiated by short-range beta-particles. The entire nucleus lies within one ^{3}H beta range of any part of a chromatid, and contains no sizable contiguous "insensitive" volume of a radius exceeding the effective range of ^{3}H beta-rays (1 to 2 μm) [6]. Experimental evidence for radiation early effects of tritium, mainly selective killing, growth delay or inhibition have been reported [7]. Administration of ^{3}HTdR to newborn mice or embryos appeared to label animals effectively for their entire life span.

Because of these unique features, the carcinogenic potency of tritium deserves a particular attention, as the site of incorporation and the selectivity of nuclear irradiation leads to speculation on possible mechanisms of action. Data available at the present time for the long-term toxicity of tritiated thymidine, with special reference to carcinogenicity, are scarce and controversial. Baserga et al. [2], observed an increased incidence of tumors and a shortened life span in CAF$_1$ newborn mice. The work of Cottier et al. [8] and Johnson and Cronkite [9] is an interesting contrast: administration of tritiated thymidine to mature BNL Swiss albino mice had no measurable effect on life span or age-specific mortality rate and did not alter the incidence or time of onset of neoplasia.

On the other hand, the biological half-life of tritiated water in mice was estimated to be from 24 to 36 hours [10,11] assuming only one biological compartment or pool. However, further studies [12-17] demonstrated that there is more than one biological compartment or pool following the consumption of tritiated water by man and animals. There are at least those associated with fatty acids, proteins, cholesterol and the nucleic acids and possibly other cellular components. It thus appears that intracellular DNA will incorporate tritium from tritiated water. This aspect of potential carcinogenicity of tritium deserves further investigation in view of the increased release of tritiated water from nuclear power reactors in the future.

Finally it is generally assumed that the nucleus is the site of the primary event initiating the carcinogenic process following irradiation. Thus, from the theoretical as well as practical viewpoint, it seemed desirable to investigate further the potential carcinogenicity of tritiated thymidine, particularly in extending the dose range to lower values.

In preliminary communications [18,19] we reported that tritiated thymidine, injected postnatally within a dose range of 0.3 to 1.5 μCi/g, significantly increased the overall tumor incidence in C57 Black/6 mice. The data available at that time were derived from gross autopsy findings. We now have completed the microscopic examination of all tissue samples collected at autopsy and the present report is based on our pathology findings.

In this experiment, all mice were allowed to live their entire life span in an attempt to collect survival data which could be used for actuarial studies on mortality. The price paid for this additional information was that, in a few instances, tissue samples for adequate microscopic study were unavailable on account of cannibalism or postmortem change, despite the fact that cages were regularly checked for dead animals, twice during working days and once during weekend days. As a consequence, the numbers of mice within various groups are slightly at variance with those previously reported.

Material and Methods

Experimental groups

Approximately 1,400 newborn C57 Black/6 mice of either sex were used. Mice were obtained from our own breeding colony which originated from pathogen-free breeders purchased in 1963 from T. C. Kile, Cumberland View Farms, Clinton, TN. In each litter, animals were randomly assigned to a given treatment group or the corresponding control group. After weaning, mice were kept in plastic cages, in groups of four to seven, with *ad libitum* access to water and Purina laboratory chow, under controlled heat and light conditions. Littermates assigned to experimental groups received tritiated thymidine at one of the following dose levels: 0.3, 0.4, 0.6, 0.9, or 1.5 μCi/g of body weight. Littermates assigned to control groups received cold thymidine. The specific activity of the tritiated thymidine used was 3.3 Ci/mmole. All injections were given subcutaneously in the suboccipital region, in the newborn mice within 16 hours following birth. The required amount of tritiated or cold thymidine was dissolved in sterile water and injected in an adjusted volume of 0.1 ml. A drop of collodion was immediately applied on the punctured skin after the injection needle was removed, in an effort to seal the wound and prevent leakage of any injected material.

Table 1 shows the experimental design and the number of mice with full pathological records and having reached adult life. Routine autopsies did not include examination of the brain. Sample tissues were routinely taken from the following organs: thyroid, lung, thymus, liver, spleen, kidney, spine and any relevant lesion that was grossly observed. The tissues were fixed in Bouin's solution and processed by routine histological procedures.

Among all injected mice, only 862 reached adult life, as a number of newborn animals were victims of cannibalism following injection

Table 1

Experimental Design

Sex	Dose μCi/g	Full Pathology Records
♂	0.3	56
♀		40
♂	0.4	55
♀		44
♂	0.6	21
♀		22
♂	0.9	31
♀		24
♂	1.5	36
♀		47
♂	Control	178
♀		205

of either tritiated or cold thymidine, and were observed throughout their entire life span, up to 1,200 days. Complete pathology records finally were collected on 759 mice.

Classification of tumors

Tumors observed in animals were histologically confirmed and divided into four groups: (1) thymic lymphosarcomas, or mesenteric lymphosarcomas or generalized tumors of the hematopoietic system involving nodes, spleen, liver, bone marrow and sometimes other organs, (2) reticulum cell sarcomas, separately tabulated as they have been shown not to be radiation induced, (3) hepatomas, (4) miscellaneous tumors of which most frequently observed were fibrosarcomas from various sites and tumors arising from the thyroid gland, the ovary and the lung.

Statistical methods

Actuarial statistics were applied to analysis of tumor incidence in experimental and control groups. Age-specific incidence rates for specific tumors were derived in the following manner. The incidence rate for a given tumor, r_s, is:

$$r_s = \frac{1}{t_i} \ln N_i/(N_i - n_i)$$

where n_i is the number of animals exhibiting a specific tumor in age interval i, of t_i days, and in a population of N_i animals at risk at the beginning of the interval. The use of natural log rates yields the best estimate of instantaneous rates of incidence, as if they were derived from an infinitely large population [20]. Significant differences were computed at the 0.05 level, using the chi-square test.

Results

Lymphosarcoma

It was arbitrarily decided that lymphosarcomas would include entities that are commonly called lymphoma, lymphomatosis and leukemia. It is quite apparent that lymphosarcoma has a leukemic aspect when one observes a large number of such diseased mice. Frequently one can find in small venules large numbers of primitive lymphoid cells. Most of the lymphosarcomatous mice examined histologically had the disease in an advanced stage.

We saw only rarely a thymus that could be remotely considered to have an early preleukemic atrophy of the thymic cortex or a focal multiplication of primitive lymphoid cells. The earliest lesion was a blurring of the cortical-medullary junction and the disappearance of the normal architecture. This, too, was relatively uncommon. The most usual lesion was an enlarged organ with a marked lymphoid hypertrophy. There was a uniform sheet of lymphoblasts and mature lymphocytes with some pyknotic cells, and a few megakaryocytes and foci of erythroblasts.

The spleen in the lymphosarcomatous mice was often a sheet of immature and mature lymphocytes which overran the white pulp. The only area spared was the amyloid deposition in the reticulum cell halo. Amyloid could at times be so extensive, however, that the lymphoid elements had little space to reside. In some cases there was a lymphoid hyperplasia even in an atrophic spleen. In such cases lymphoid hyperplasia was usually observed on one or more lymph nodes, i.e., mediastinal, pulmonary, perirenal, inguinal, etc.

Lymphoid hyperplasia was less common in the liver, kidney, adrenals and lungs but could be quite extensive and at times displace considerably the parenchyma. Reticulum cell hyperplasia did occur independently or concurrently with lymphoid hyperplasia. The reticulum cells most commonly formed nodular masses in the liver, kidney and spleen that appeared to arise from reticuloendothelial centers.

Hepatomas

Hepatomas ranged from small nodules that escaped detection at autopsy to large overwhelming masses that obscured the normal structure of the liver. The small tumors usually were seen microscopically as sharply delineated groups of cells which would interrupt the usual pattern of the liver cells. The tumor cells were often not greatly different from the hepatocytes except for their enhanced size but did show some degree of pleomorphism. The large tumors on the other hand had a loss of lobular pattern and rarely, if ever, could portal triads or bile ducts be observed. The blood supply of the tumor of the large variety was apparently from hepatic arterioles. Some tumors were composed of strands or cords of tumor cells separated by large blood-filled pools. Others were large sheets of thickened cords. Still others were composed of tube-like structures with variably sized bile canaliculi. No bile pigments were seen in these tubes.

Amyloid was on occasion seen in the tumor, most particularly if the deposits were also seen in the normal parts of the liver. In addition, lymphosarcomatous collections in the tumor mass would often be seen. Proliferation of bile ducts was quite uncommon and never severe in extent.

Thyroid

A most common finding was a microfollicular hyperplasia in which the follicular epithelium was very tall but usually no colloid was formed. Thyroids with this change were far more numerous than normal thyroids. A few cases were seen in which the follicular epithelium was very tall and much colloid with scalloping was found. Rarely was there papillary infolding or dumbbell shaped lumina. Also observed on a few occasions was exhaustion atrophy with loose, diffuse fibrosis of the interfollicular tissue and desquamation of epithelial cells into the thin colloid of the lumina. Occasionally amyloid deposits were observed in the vascular interfollicular spaces and they were always associated with some atrophy of the follicles.

Bone marrow

Sections were from the lumbar spine. In the younger animals, the myeloid and erythroid cells were predominant but, with advancing age and a much higher tendency for lymphosarcoma to develop, the lymphoid cells were usually equal to and frequently far greater than the myeloid and erythroid cells. In rare instances, the lymphoid cells had essentially crowded out all but a few megakaryocytes and reticuloendothelial cells.

Lungs

Pulmonary edema was a common finding. It is believed that it was an agonal or post-mortem phenomenon since very few of the mice were killed, but were permitted to die.

Nearly all of the old mice had acute, and sometimes chronic interstitial pneumonitis, often with atelectasis. Focal abscesses were infrequently found and bronchopneumonia was equally rare except in the most advanced disease. In the younger animals, the lesion was locally diffuse and located in the alveolar septa. Neutrophiles, plasma cells, macrophages were the common cells. They were hardly ever found in the alveola. In the advanced disease, the bronchii were dilated, often with inflammatory exudate, and atelectasis was much more prominent.

The common tumor of the lung observed was the pulmonary adenoma which arises from alveolar septa. Most were seen at necropsy but a few were only found microscopically. Often there were many small focal tumor masses. Less common was adenomatosis. There was metaplastic transformation of the alveolar lining cells into cubiodal or columnar types with a clear lightly staining mucoid substance in the alveoli.

Metastatic masses from known tumors were most uncommon in the lung. In one case it was clearly from a hepatoma. Amyloid was occasionly observed in alveolar septa but never in tumors. It was most easily seen at or near the tips of the lobes.

Kidney

Glomerulosclerosis or glomerulonephritis were frequently seen. It was as might be expected most frequent in the older mice. The glomerulosclerosis in its early manifestation as a glomerulitis was confined to the mesenchyme but, as the disease progressed with age, the entire glomerulus was involved with the hyaline deposition. In glomerulonephritis cases, as the disease advanced there was endothelial proliferation and adhesions of the glomerclus in one or many portions of the glomerular capsule. The tubules would usually be dilated and filled with a faint staining material in the later stages.

Polycystic kidneys, or possibly hydronephrotic, were a fairly common finding. Less common was interstitial nephritis and on occasion pyelonephritis.

Tumor incidence rate

Tables 2 and 3 give the overall incidence rates of tumors in pooled female and pooled male control mice, as well as their respective median survival times. It is seen that the "spontaneous" overall incidence of tumors was very high in both sexes and comparatively higher in females than males. Whereas the crude incidence of lymphosarcomas was higher in females, there was a suggestion that liver and thyroid tumors were more numerous in males. Some unusual tumors were found such as an adrenal adenoma and a prostatic adenoma, both in male animals. Also multiple tumors in a single animal were frequent as evidenced in comparing the total number of tumors and the number of tumor-bearing animals, both in males and females.

Figure 1 shows the age-specific tumor incidence rates in control male and female mice. Those rates were not significantly different in either sex. Tumor incidence rates appear linearly related with age up

Table 2

Overall Incidence of Tumors in 205 ♀ Mice

	Number	Percent	Median Survival
Lymphosarcoma	112	54.6	743
Reticulum Cell Sarcoma	5	2.4	809
Liver	10	4.9	763
Thyroid	10	4.9	769
Fibrosarcoma	7	3.4	831
Ovary	5	2.4	729
Lung	4	2.0	816
Others	7	3.4	646
Total of Tumors	160	78.0	

Table 3

Overall Incidence of Tumors in 178 ♂ Mice

	Number	Percent	Median Survival
Lymphosarcoma	60	33.7	721
Reticulum Cell Sarcoma	4	2.4	670
Thyroid	19	10.7	701
Liver	15	8.4	747
Lung	9	5.1	745
Others	5	2.8	721
Total of Tumors	112	63.0	

Figure 1

Age-specific Tumor Incidence Rate in Control Male and Female Mice

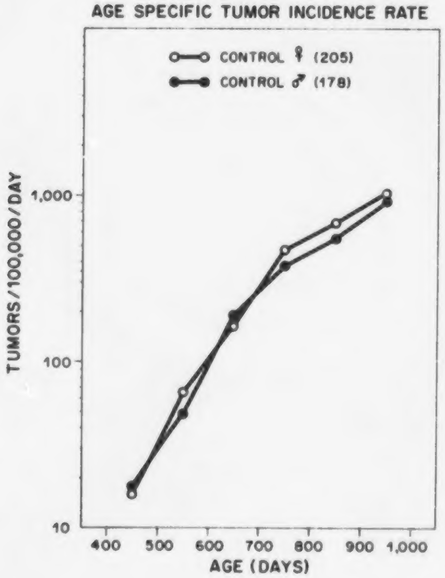


Figure 2

Age-specific Incidence Rate for Tumors of Reticular Tissue (Thymic, Mesenteric and Other Lymphosarcomas, Exclusive of Reticulum Cell Sarcomas) in Males and Female Mice

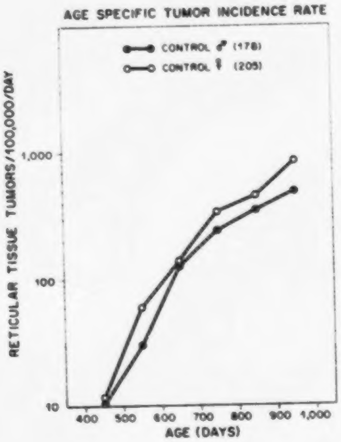


Figure 3

Age-specific Incidence Rate for all Tumors in Male Controls Versus Male Experimental Animals Injected with Tritiated Thymidine at any Dose Level

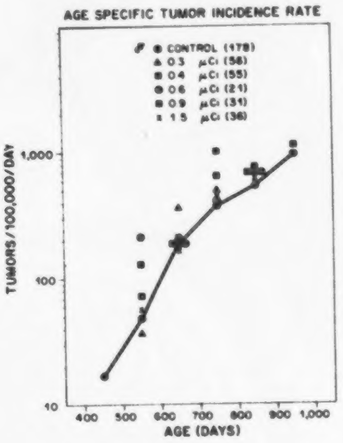


Figure 4

Age-specific Incidence Rate for all Tumors in Female Controls Versus Female Experimental Animals Injected with Tritiated Thymidine at any Dose Level

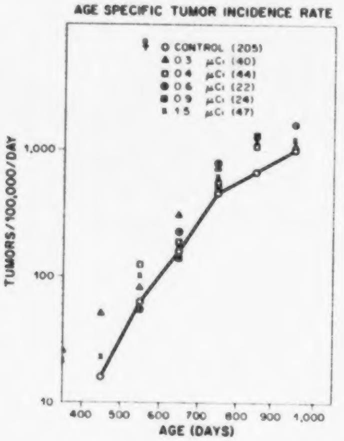


Figure 5

Age-specific Tumor Incidence Rates for Reticular Tissue Tumors in Male Control Versus Male Experimental Animals Injected with Tritiated Thymidine at any Dose Level

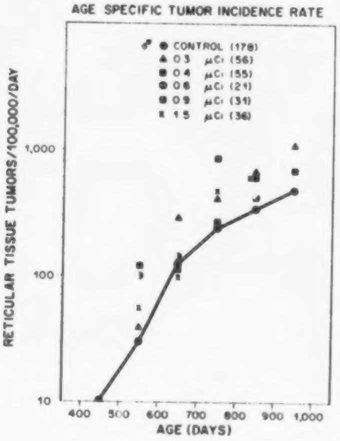


Table 4

Tumor Incidence in Tritiated Thymidine ♀ Recipents (177)

	Number	Percent	Median Survival
Lymphosarcoma	117	66.1	736
Reticulum Cell Sarcoma	2	1.1	754
Liver	8	4.5	735
Thyroid	6	3.3	746
Fibrosarcoma	5	2.8	876
Ovary	2	1.1	791
Lung	6	3.3	760
Others	4	2.2	752

Table 5

Tumor Incidence in Tritiated Thymidine ♂ Recipients (199)

	Number	Percent	Median Survival
Lymphosarcoma	81	40.7	700
Reticulum Cell Sarcoma	1	0.5	624
Liver	16	8.0	731
Thyroid	20	10.0	689
Fibrosarcoma	2	1.0	554
Testis	2	1.0	615
Lung	8	4.0	736
Other	2	1.0	785

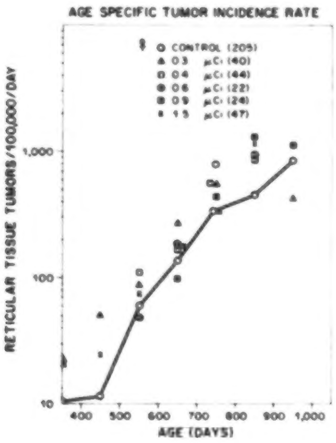
Table 6

Variation of Tumor Incidence in Percent
In Tritiated Thymidine Recipients (376)

	♀ Mice	♂ Mice	P
Lymphosarcoma	+11.5	+7.0	<0.05
Thyroid	- 1.6	-0.7	
Liver	- 0.4	-0.4	
Lung	+ 1.3	-1.1	
Others	- 1.2	-1.8	
All Tumors (Excluding reticulum cell sarcomas)	+ 7.3	+4.9	<0.05

Figure 6

Age-specific Tumor Incidence Rates for Reticular Tissue Tumors in Female Control Versus Female Experimental Animals Injected with Tritiated Thymidine at any Dose Level



to 750 days. Thereafter, inflection of the curve indicates a discontinuity in the rate increase of tumors.

Figure 2 shows the age-specific incidence rates for tumors of reticular tissue (thymic, mesenteric and other lymphosarcomas, exclusive of reticulum cell sarcomas), in male and female animals. As expected, incidence rates were higher in females than in males throughout life span.

Figure 3 shows the age-specific incidence rates for all tumors in male control versus male experimental animals injected with tritiated thymidine at any dose level. It is seen that most representative points are plotted (with exception of three) in the area above the control curve, i.e., they indicate higher age-specific rates for tumor incidence. Small sample sizes may be responsible for the noted discrepancy: the three aberrent points represent respectively 2, 2 and 2 lymphosarcomatous animals.

Figure 4 shows the age-specific incidence rates for all tumors, in female control versus female experimental animals, injected with tritiated thymidine at any dose level. Again it is seen that, with the exception of two representative points, all others fall above the control curve, thus indicating higher age-specific rates for tumor incidence. The two aberrent points are once more representative of small sample values, respectively 1 and 2 lymphosarcomatous animals.

Tables 4 and 5 show the incidence of various tumors, respectively in female and male recipients of tritiated thymidine. It is seen that the relative increase of lymphosarcoma incidence as a result of irradiation was more pronounced in females than in males. On the other hand, there was a suggestion that tumors of liver and thyroid were relatively less numerous in females than in males. Similarly the median survival time for either tumor was lowered in males.

Table 6 shows the variation of tumor incidence in both male and female recipients of tritiated thymidine versus control animals. It is seen that the overall tumor incidence was similarly increased in both sexes. The observed increments were statistically significant by chi-square test at the 0.05 level. The overall increase in crude tumor incidence appeared to be attributable to enhanced lymphosarcoma incidence. Fluctuations in incidence of other tumors were not statistically significant.

Figures 5 and 6 show the age-specific tumor incidence rates for tumors of reticular tissue, respectively in male and female controls versus animals injected with tritiated thymidine. Most incidence rates for injected mice are higher than corresponding control rates.

No statistically significant dose-response relationship could be derived for tumor incidence rates, between different dose levels in groups of mice injected with tritiated thymidine, mainly because of small sample sizes.

Discussion

The final results of this experiment confirm and extend our previous observations [21,18,19]. They indicate that tritiated thymidine, when injected postnatally within a dose range of 0.3 to 1.5 μCi/g significantly increases the overall tumor incidence in C57 Black/6 mice. The increased incidence of lymphosarcomas was, in both sexes, the main factor contributing to the observed overall tumor incidence. For other types of tumors, incidence appeared either unchanged or even sometimes decreased. For tumors observed within samples of sufficient size, median survival times of tumor-bearing animals were shortened following administration of tritiated thymidine. However, variation of tumor incidence could not be attributed to differences in life span, as variation of age-specific incidence rates actually paralleled the crude incidences. Thus, in this experiment, the phenomenon of alteration (i.e., enhancement or inhibition) of the spontaneous, normal carcinogenesis, concurrent to the presence of tritiated thymidine in the cell nucleus, could not be accounted for as a mere time variant.

The present data confirm for the C57 Black/6 strain the observations of Lisco et al. [22], establishing that various types of malignancies may be induced in CAF_1 mice following administration of tritiated thymidine in doses of 1.0 μCi/g. They further indicate that smaller doses are effective in increasing the cancer incidence. The carcinogenic potency of a given dose may possibly depend upon the age of recipients, as in the experiment of Lisco et al., mice were injected from two to seven days after birth, whereas in our study injection was carried out within 16 hours following birth. Dependence of susceptibility upon age is further suggested by the negative results obtained by Johnson and Cronkite [9]. They found no change in age-specific mortality rates nor alteration of incidence or time of onset of neoplasia in mature BNL Swiss albino mice injected with tritiated thymidine, in doses of 1 or 5 μCi/g of body weight, at ages varying from 6 to 12 weeks. The latter results are at variance with the observations of Lisco and Baserga [22] as well as with our own, and possibly reflect a difference in susceptibility. Also, the divergence of findings might result from strain variations in turnover rates and pool sizes, affecting the amount and distribution of the residual activity of tritium throughout the life span of animals, and consequently altering the parameters of dosimetry on macroscopic and microscopic scale.

The carcinogenicity of tritiated thymidine in mice lends itself to speculations on its mode of action. The main features of ^{3}HTdR incorporation in living tissues have already been pointed out, namely the selective irradiation of the nucleus, the dependence of intensity of labeling upon cell population dynamics in various organs, and the permanence of residual activity in the cell genome.

The pattern of tumor appearance and distribution in control animals is strikingly suggestive of a stochastic process associating a definite frequency with a given type of tumor. The multiplicity of tumors arising in a single animal and the fact that the increased frequency of one type of tumor (as elicited by tritiated thymidine) is concurrent to a decreased frequency of one or more other types, further suggest that some competitive process may take place during the emergence and development of such tumors. Also the linearity of the age-specific rates with age, as expected from a true Gompertzian regression, would meet the assumption that the carcinogenic process is dependent upon two interacting component mechanisms, one of phenotypic expression, acting at a constant rate of intensity throughout life span, and one of repression, gradually losing strength, (i.e., acting as a derepression) also at a constant rate throughout life span. It is very tempting to visualize such a dual mechanism as one of "turning on and off" genes, most particularly oncogenes.

The high frequency of spontaneous tumors in pooled control animals, averaging about 70% in our experimental conditions, is highly suggestive of the permanence of oncogenes in most, if not in all C57 Black/6 mice. It might be that with further extended longevity, the 100% landmark could be approached more closely. As a consequence, cancer should be thought of as a normal event, directed from genetic information previously stored in the cell genome in a "turned off" state. Tritiated thymidine would no longer appear as an inducing but rather as a triggering agent, "turning on" oncogenes with greater probability, thus counteracting intrinsic mechanisms of normal genetic derepression.

Finally the tumor data in this experiment and their implications point out that observation of the animals during entire life span is an absolute prerequisite for the proper design and interpretation of experimental oncogenesis in mammals.

Summary

The carcinogenic potency of tritium at low dosage is controversial. The need for additional data is urgent in view of the increased release of tritiated water from nuclear power reactors in the forseeable future. A total of about 1,400 newborn male and female pathogen-free C57 Black/6 mice received either tritiated thymidine subcutaneously at one of the following dose levels: 0.3, 0.4, 0.6, 0.9, and 1.5 µCi/g of body weight, or cold thymidine (controls). Due to cannibalism and other causes of early mortality, 759 mice were observed through entire life span up to 1,200 days and were routinely autopsied at death. Microscopic studies of selected tissues were performed on all animals. The overall tumor incidence for pooled control groups was found to exceed 70%. Age-specific incidence rates appear to fit a linear Gompertzian regression up to 750 days of age for all tumors, including thymic lymphosarcomas, lymphomas, leukemias, hepatomas, thyroid and lung adenomas, fibro- and osteosarcomas, adenocarcinomas of the G.I. tract, etc. In groups of animals treated with tritiated thymidine, age-specific incidence rates for all tumors were higher in most instances. It is concluded that tritiated thymidine injected postnatally within a dose range of 0.3 to 1.5 µCi/g significantly increases the overall tumor incidence in C57 Black/6 mice. Furthermore tritiated thymidine appears to initiate a triggering rather than an inducing process in tumor production.

ACKNOWLEDGMENTS

This work was supported in part by Contract 020-62-12 BIOB, Euratom, Universite Libre de Bruxelles, Contract 396/R/1/RB, International Atomic Energy Agency, and NIH General Support Grant, PHS 5 S01 RR05367.

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TWO CASES OF TRITIUM FATALITY

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It is obvious that tritium is quite an interesting nuclide, for a number of different reasons:

Tritium is an isotope of an element that is very common in the crust of the earth and is present in all organic compounds. It is, and increasingly will be, present not only in water but in all living matter. This last fact has not always been taken into account in the conventional models for the turnover of tritium and for the effects on the living organism exposed to radiation from this source. In particular, the possibility of the nonuniform distribution of tritium in living matter has not been considered carefully enough. A microscopically nonuniform distribution of tritium, with its very short-range beta-radiation may influence the biological effect considerably.

Physico-chemically, tritium is particularly interesting in that its mass differs not by only a few percent from that of its nonradioactive isotope, as with most radioisotopes, but by a factor of 3. The isotopic effect as discussed by Weston [1] and possibly other effects, leads to changes in shape or form of a tritiated molecule or compound and, consequently, changes in its physico-chemical behavior. These factors may have more serious consequences than in the case of other radioisotopes. To what extent such theoretically postulated effects may influence, for example, the reduplicating process of DNA is not completely known. This is of particular interest since, quantitatively, the isotopic effect depends on the number of atoms present and not merely on the number of disintegrations per second. In the case of tritium, with its long physical half-life, the number of atoms present is 560 million times the number disintegrating per second, and for each ion pair produced by the radiation from tritium, about three million atoms have to be present.

As a consequence of its physico-chemical characteristics, it is a property of tritium that, where atomic exchange occurs, the transfer velocity differs from one chemical compound to another. This means that where the concentration in the water decreases, the concentration in the organic compound can remain at a much higher level for a long time. This is particularly important where an estimation of the body burden is based on measurements of tritium concentration in urine; in such cases it can be extremely difficult to achieve a realistic judgment.

Tritium is interesting, in addition, because it is not only produced in large amounts as waste in modern nuclear technology but also is used in industry and research. Let us take, as an example, the Federal Republic of Germany where, in 1968, nearly all the radionuclides used were imported, so that the import statistics give an exact picture of the usage. In that year a total of 28.8 kCi of radioisotopes were imported, excluding sealed sources of activity greater than 50 Ci, e.g., Cobalt-60 sources for uses in radiotherapy. Of this imported radioactivity, 56% was tritium and 34% Iridium-192 in sealed form. If we consider only the unsealed radionuclides, deducting iridium from the total amount and assuming that all the others, including Cobalt-60 sources of less than 50 Ci were unsealed, the total imports of unsealed radioisotopes amounted to 18.9 kCi, and tritium accounted for 85% of this activity. No definite figures are available for the total tritium activity introduced into luminous compounds and of the activity lost during this process. Both together may exceed 50% of the total imported activity, 16.1 kCi of tritium in the year 1968.

Finally, tritium is interesting because in the past it was difficult to measure, and everybody is familar with the concept that "A hazard that is not measurable is not regarded as existing." It was not surprising, therefore, a few years ago to see on a bottle containing tritiated luminous compounds, a label bearing the statement: "Tritium: nonradioactive."

The aim of this paper is not just to tell a case history. Rather, it is to show the practical problem and to stimulate further investigations and considerations.

The cases being reported are those of two persons who died and of a few who became ill, having worked with tritium in activities of 50 to 100, up to a maximum of 1000, Ci per batch, dealing with a total activity of 1000 to 3000 Ci per year for 3 to 4 years. These cases occurred in different countries in Europe. There is no absolute proof that the fatalities were caused by tritium. However, the similarity of both cases with regard to circumstances, clinical and radiological case histories, clinical symptoms similar to those reported in other cases of radiation fatalities (such as that of Madame Curie), and radiation doses in excess of the maximum recommended by ICRP, are proof, to the author, of a causal relationship.

One case has been published by Minder [2], and a few additional details shall be given here; the other case has not yet been published. In both cases, factories had been engaged for years in manufacturing luminous compounds for dials, etc., activated with radium. Later on, they worked with promethium and strontium and, a few years before the cases occurred, with tritium.

Tritiated luminous compounds are prepared by hydrogenating the basic materials for certain plastics with tritium, polymerizing these tritiated compounds, dissolving the polymerized tritiated plastic material in organic solvents, mixing the solution with fluorescent zinc sulfide crystals, and finally evaporating the solution. The end product, i.e., the crystals, are covered by a thin layer of this plastic material. During all these procedures, tritium is evaporated as gas or vapor, and even the end product and the painted dials lose tritium. Depending on the quality and the specific activity, up to 50% of the tritium initially present can be lost during the first few months.

Work with tritium, in both the cases to be described, lasted from three to six years with increasing activities being used, ranging from less than 100 up to several thousand Ci/year. Urine samples taken in the later periods of work showed concentrations of 2.2 to 49 times the maximum permissible concentrations (MPC) in urine, derived from the ICRP recommendations, on the basis of the conventional assumption of 1 mCi maximum permissible body burden for the soft tissues or body water, amounting to 63% of the body weight. Furthermore, it is assumed that tritium is uniformly distributed in the body and the MPC is calculated as 23 nCi/ml urine. According to other methods of calculation, it amounts to 29 nCi/ml, which is practically the same.

It should be mentioned in this context that Bond [1] considers the ICRP values as too conservative with a factor of 5 to 8 and an average concentration of 150 to 200 nCi/ml urine would in this case correspond to a whole body exposure of 5 rem/year.

In both cases, the persons concerned died with the clinical signs of an aplastic Panmyelocytopenia complicated by pulmonary or other symptoms. Both showed certain clinical symptoms intermittently, e.g., nausea, lassitude, exhaustion, etc. Also, blood counts after one to two years' work with tritium showed only a slight and variable decrease in the white cells, without significant abnormalities with regard to type, but a continuous decrease in the red cells, though less so in the hemoglobin, in other words, a hyperchromic type of progressive anemia. In the following accounts, more details of the two actual cases are given and the measured data presented.

Case A

In Case A the main person involved, "A.1," was an engineer who was 60 years of age when he started to work with tritium and who had worked since 1937 with thorium and radium in the construction of measuring equipment, etc. From 1955, he was engaged in research on radioactive luminous compounds using beta-emitters, and in 1957 he started to work with a few Ci of tritium as gas and water. In 1958, he obtained a license to work with a total of 600 Ci, and in 1959 a license to work continuously with 500 Ci and to store 2500 Ci, which was increased in 1963 to 1000 and 3500 Ci, respectively. However, he never used more than 150 Ci per batch, and in the final period about one batch per one to two months. The estimated consumption of tritium was about 600 Ci for 1957-60, 2500 Ci for 1961, 2300 Ci for 1962, 1000 Ci for 1963 and 1150 Ci for 1964; altogether, therefore, about 7.5 kCi over eight years and about 7 kCi over four years.

Reliable methods for continuously monitoring tritium were only in the development stage during the first years of this work. Personnel monitoring with film badges, however, showed "high doses of hard radiation" obviously caused by diffusion of tritium into the film.

In December, 1962, the monitoring of urine samples was started and during 1963 (about 700 to 350 days before death) showed tritium concentrations of 140 up to 1120 nCi/ml or about 6 to 50 times the MPC for urine of 23 nCi/ml. One value of more than 2000 nCi/ml is not considered, as a value of only about 1000 nCi/ml on the following day indicated contamination of the vessel or some other error in the measurements. In 1964, concentrations of about 50 up to a single maximum of 800 nCi/ml, or 1.3 to 35 times the MPC, were found.

Figure 1 shows the tritium concentrations as measured for the subject "A.1" and his technician "A.2" (dotted lines). The abscissa indicates the number of days before (-) or after (+) the work with tritium was stopped thus giving the possibility of better comparison of the different cases. The biological half-life of 12 days, as considered by ICRP, is shown for optical comparison. The figures are listed additionally in Table 1. Also the activity of tritium

Figure 1

Tritium in Urine
Cases A.1 and A.2

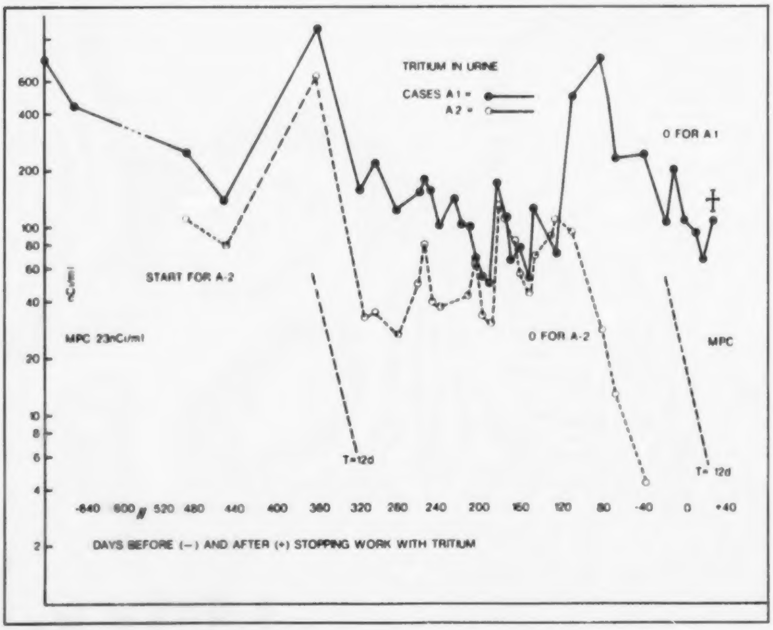


Table 1

Tritium Concentrations in Urine in Two Cases of Tritium Fatalities

Concentrations in nCi/ml; Time in years/days before (-) and after (+) ceasing work with tritium; * = persons who died; "Activity" in Ci used for a single hydrogenation (H), or bought once (B), or licensed for continuous use (L) or for storage (S); Figures with ? suspect for contamination of vessel.

Person A.1*		Person A.2		
time	urine	time	urine	activity
-7/158	start	work with	^{3}H	B 1
-6/315				B 30
		-6/132	appointed	
-6/154				B 100
-5/203				B 500
-5/ 14				L 500
-3/200				S 2500
-1/312	820		-	
-1/284	465		-	
-1/153	-		-	L 1000
-1/116	258	-1/10	110	
-1/ 81	140	-340	80	
-356	2360?	-250	640?	
-355	1120		-	
-313	155	-207	32	
-302	216	-196	34	
-280	123	-174	25	H 50
-266	-		-	H 50
-259	152	-153	50	
-252	181	-146	85	
-245	158	-139	40	
-238	103	-132	- 36	
-231	-	-125	183	H 100
-224	145		-	
-216	104		-	
±212	-	-106	43	
-210	102		-	
-203	68	- 97	67	
-196	55	-90	34	
-189	51	-83	31	
-182	173	-76	133	H 100
-175	115	-69	114	
-168	69	-62	88	
-161	77	-55	57	
-153	55	-47	45	
-147	127	-41	71	
-134	-		-	H 100
-133	-	-27	90	
-126	76	-20	111	
-112	512	- 6	95	H 150
-106	-	0	stop of	work
- 84	795		-	
- 83	-	+23	28	
- 77	500		-	
- 70	236	+36	13	
- 63	-		-	H 150
- 56	-		-	H 150
- 42	247		-	
- 41	-	+65	4.5	
- 21	108	+85	2.3	
- 13	206	+93	2.0	
- 8	111			
0	since stop of work			H 100
+ 7	95	+113	4.1	
+ 14	70		-	
+ 24	110			

licensed or used is mentioned.

A reliable and accurate estimation of the average concentration is impossible as no measurements of urine concentrations are available for the initial period, and an estimation of concentrations between measurements is extremely difficult. Integrating the area below the curve and considering the activity used during the time when no measurements were available, an average whole body dose of 19 rem within the first two years and 93, 85, 37, and 34 within the following four years, respectively, with an overall dose of 287 rem, were calculated. The estimated maximum possible (but improbable) value of the extrapolated dose is 541 rem.

The extrapolation was done for the last 400 days, with the assumption that immediately after a measurement an incorporation occurred leading with a T_b of 12 days to the concentration measured at the next measurement. With this maximum possible incorporation for this year, and taking into account the type of work and activity used, the mentioned "extrapolated" figures were derived.

The above-mentioned radiation doses were calculated according to the ICRP model, which takes into account the tritium content of the body water only. A simple two-compartment model, however, developed by Gottschweski [5] considering results from other experiments not reported here and the long-term values of Minder's case, shows that 1 to 2% of both a single or chronic intake of tritium may be transferred to the organic compounds of the tissues.

The model is expressed as the sum of two exponential functions with biological half-lives of 8 to 12 and 90 to 120 days, respectively. Theoretically, of course, one may have to assume as many compartments, and therefore exponential functions, as biochemical compounds exist in a body. For practical purposes, however, a minimum of two and a maximum of four may be sufficient. In the case of chronic intake, equilibrium will be reached in about 20 to 30 days, when the concentration in organic matter will be about 12% of that in body water. After stopping the intake, the absolute values of tritium concentrations in body water and tissue will decrease, but with different rates, and the tissue concentration relative to that in body water will increase. Seventy to ninety days after stopping the intake, tritium concentrations in organic material are expected to be 8 to 16 times as high as that measured in urine. Assuming a uniform distribution of this tritium also, the whole body dose due to this component is estimated to be 7 to 14% of the body water dose. Almost 300 or extrapolated 600 rem, most of which was absorbed within the last four years of work, particularly in 1962 and 1963, would then be the result. This dose would still be in the order of magnitude of the "Tolerance dose" accepted 35 years ago.

Clinically, case A.1 showed, over a number of years, certain symptoms of hypertension, duodenal ulcer and hyperthyroidism. The blood counts were normal, however. It was only after January 1964 that symptoms of a general decline in health, with lassitude, nausea, etc., occurred and were initially ascribed by the medical practitioner concerned to the diseases already diagnosed.

From November 1961 onwards, when blood counts were still in the normal range, until his death in November 1964, a continuous decrease in the red cells could be seen, associated with a similar but smaller decrease in the hemoglobin, whereas the white cells were not significantly changed. It was not until a few weeks before his death that they showed a significant decrease. The figures are given in Table 2.

The subject, A.1, rejected all advice from his physician, failed to follow the instructions of the government authority, and continued to work until October 14, 1964. On November 3, he was admitted to a hospital and died on November 7 after having been given a blood transfusion.

The autopsy showed an advanced Panmyelophthisis, with a marked reduction in bone marrow cells, immature forms, etc., as the immediate cause of death. In the duodenum, only slight scars due to ulceration were seen, which certainly could not have been the cause of chronic bleeding.

The tritium concentration at the time of death, in the urine and other body fluids, was 110 nCi/ml. In the dehydrated organic material, average concentrations of 24 nCi/g in the bone marrow and 25 nCi/g in the testes were found (although with variations in the results of individual measurements by a factor of 3, possibly the result of loss of activity due to the method used). The figures are quoted in Table 3. Although the figures for the tritium concentration in organic material are somewhat uncertain, they agree fairly well with the above-mentioned model,

Table 2

Blood Pictures in Two Cases of Tritium Fatalities

Time in years/days before (-) and after (+) ceasing work with tritium
Hb_E = hemoglobin per erythrocyte; sc = stab cells; Po = polymorphs;
Ly = lymphocytes; Mo = monocytes; eo = eosinophils; Ba = basophils

Time	Hemo-globin g% = %	Erythro-cytes $10^6/mm^3$	Hb_E $\gamma\gamma$	Leuko-cytes mm^3	Throm-bocytes $10^3/mm^3$	Sc %	Po %	Ly %	Mo %	Eo %	Ba %
Case A.1: Start work with tritium: Time -7/158											
-2/346	15=94%	4.6	32	3600	276	6	48	38	5	3	-
-1/178	13=81%	3.8	32	3800	208	7	39	48	4	2	-
-342	12.6=79%	4.0	33	5200	240	3	74	18	4	1	-
-204	12.5=78%	3.7	34	4100	203	8	39	40	7	3	2
-124	11.5=72%	3.6	32	6400	210	3	50	40	4	3	-
- 36	10.1=63%	3.2	31	3500	140	--	56	40	1	3	-
- 23	9.0=56%	2.6		4100		↑					
+ 16	5.8=36%	1.35		2250			"not abnormal"				
+ 24											
Case A.2: Start work with tritium: Time -6/132											
-2/194	12=75%	3.7	33	3700	215	3	48	42	5	2	-
-1/349	12=75%	3.7	32	3000	244	6	42	48	3	1	-
-1/136	12.8=80%	3.8	34	4300	190	3	53	37	6	1	-
-256	13.6=85%	4.2	32	4200	230	2	64	28	4	2	-
-151	12.2-76%	3.8	32	5000	190	5	64	23	5	3	-
-103	11.8=74%	3.6	33	3700	307	5	47	42	3	3	-
- 19	12.6=79%	3.9	32	3900	300	1	60	28	2	9	-
Case B.2: Start work with tritium: Time -3/0											
-3/160				4000			60				
-1/103	55%	2.25		950							
-1/ 76	64%	3.3		1350							
-1/ 20	79%	4.0		3200							
-314	94%	4.3		4000							
-251	90%	3.5		3500							
-136	92%	3.6		2300							
- 74	96%	3.6		2400							
- 4	92%	3.9		2500							

Table 3

The Tritium Content (nCi/ml or /g) of the Body at the Time of Death in Two Cases of Tritium Incorporation and Death by Panmyelophthisis

Case A.1:

Urine	110
Blood	110
Transudate	107

Body water extracted with xylene from

Gonads	100
Bone marrow	109

Body tissue:	Gonads	Bone marrow
Water content %	83%	21.5%
Fat content %	-	65.0
Tritium measured	24.5	23.7

(After incineration: variations between different samples, however, with a factor of 3).

Case B.2:

Tissue	Water Content %	Fat Content %	nCi/g tissue		
				calculated excluding	
			measured	water	fat + water
Urine	100	-	2.5	-	-
Muscle	73	7.2	29.0	108	146
Kidney	65	5.0	28.0	80	94
Liver	61	21.1	24.0	62	134
Spleen	67	3.1	18.0	55	60
Lung	80	1.2	14.0	70	75

NOTE: Even when the tissue was kept over months in formalin, it did not eluate any activity, which shows that the activity is in the organic material and not in the body water.

according to which about 10% of the average concentration in the body water is transferred to the organic compounds in the case of chronic intake. The average concentration over the last two years was calculated as 220 nCi/ml. Incorporation of radium, thorium, strontium and their daughter elements was found to be negligible compared with that of tritium (0.4 1 pCi Ra-226/g bone).

A second subject involved in this case, "A.2," had assisted A.1 since 1958 and stopped work in the middle of 1964. The whole body exposure, calculated from tritium concentrations in the urine, ranged from an average figure of 124 rem up to an extrapolated figure of 278 rem, mainly accumulated during the last four years of work and amounting to about one half of that for A.1.

It is of interest to note that this person, A.2, would not have exceeded the exposure calculated from the ICRP formula during the first three years of work, although she showed slight symptoms in the fourth year. She was 22 years of age when she started work with tritium and showed a moderate hyperchromic anemia during the last four years. A detailed medical report on the case is unfortunately not available.

The subjects "A.3" and "A.4" started to work after A.1 and A.2 had stopped. In the meantime, radiation protection measures had been improved and A.3, who was 61 years of age and a chemist, had tritium concentrations of 10 to 89 nCi/ml in the urine. A.4, his assistant, aged 35, had 1 to 48 nCi/ml during the following 11 months; the workshop was then closed by the government authorities. Seven months later both persons still showed 0.3 to 0.6 nCi/ml in the urine. Serious clinical symptoms were not reported in the cases of either A.3 or A.4. Figure 2 and Table 4 include the values of these subjects; values indicated with "?" are possibly somewhat too high, as there was suspicion of contamination of the urine samples with tritium.

Figure 2

Tritium in Urine
Cases A.3 and A.4

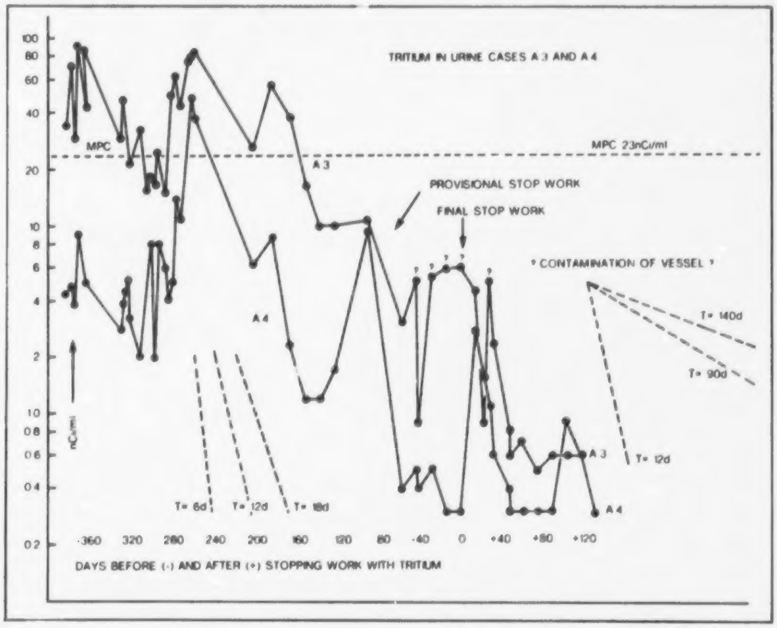


Table 4

Tritium Concentrations in Urine in Two Cases of Tritium Fatalities

	Person	
Time	A.3	A.4
-412	start	work
-382	34	4.4
-381	32	4.8
-379	47	4.2
-378	68	4.8
-377	40	3.6
-376	35	4.4
-375	29	3.8
-374	55	6.3
-373	77	10.0
-372	89	9.0
-371	80	7.0
-367	88	6.0
-364	43	5.0
-330	29	2.8
-329	46	2.6
-328	31	3.8
-327	27	3.6
-325	30	4.4
-324	28	3.6
-323	-	5.1
-322	21	3.1
-312	32	2.0
-310	19	3.0
-307	16	4.0
-302	18	8.0
-298	17	2.0
-295	24	8.0
-288	15	6.0
-285	24	4.0
-281	49	5.0
-278	61	14.0
-274	44	11.0
-27[illegible]	45	11.0
-266	75	31.0
-263	82	48
-260	84	37
-204	26	6.1
-186	56	8.5
-169	38	2.3
-154	16	1.2
-140	10	1.2
-126	10	1.7
- 94	10.4	9.5
- 60	provisionally stop work	
- 59	3.0	0.4
- 45	5.0?	0.5
- 44	0.9	0.4
- 30	5.3?	0.5
- 16	5.9?	0.3
- 2	6.0?	0.3
0	restart of work for one day only	
+ 12	4.6	2.7
+ 21	0.9	1.5
+ 26	5.1?	1.1
+ 30	2.3	0.6
+ 45	0.8	0.4
+ 46	0.6	0.3
+ 58	0.7	0.3
+ 72	0.5	0.3
+ 86	0.6	0.3
+100	0.6	0.9
+114	0.6	0.6
+127	-	0.3

	Person
Time	B.2*
-3/0	start work
-109	53
- 20	88
- 19	117
- 10	58
- 7	96
0	stop of work
0	70
+ 74	5.0
+ 96	29
+108	13
+144	38
+167	12
+187	21
+206	8.2
+212	4.3
+229	10.0
+234	4.0
+244	6.7
+276	4.3
+289	8.0
+296	9.0
+318	5.3
+346	5.0
+374	3.9
+376	3.7
+388	2.2
+391	3.4
+456	2.8
+480	2.5

Case B

In Case B, three subjects were involved, namely "B.1," the person in charge of production for many years, "B.2," an assistant employed in the factory for two years before work with tritium was started, and "B.3," who had performed administrative functions over the period that B.1 had been in charge. Figures are available only for B.2.

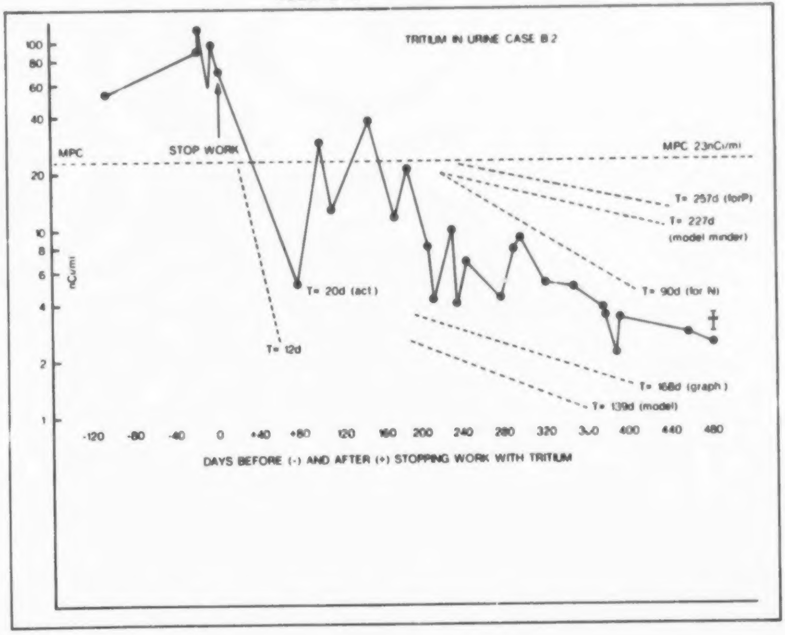
Luminous compounds had been produced in this factory for many years with radium-226 and for a few years with strontium-90. Incorporation of radium slightly exceeding the maximum permissible body burden was reported for B.1. For B.2, four years after work with radium and strontium was stopped, about 90% of the MPBB for radium was found. However, when work with tritium was started, this person still had a normal blood picture and no clinical symptoms of any radiation-induced disease.

The work with tritium involved the production of luminous compounds with 200 to 1000 Ci of tritium per batch and a total of several thousands Ci of tritium over a period of three years. Exact figures are unavailable.

About 32 months after starting work with tritium, B.2 developed symptoms of a radiation syndrome, e.g., nausea, lassitude, etc. Measurements of tritium concentrations are available for B.2 over the period from 18 months after starting work with tritium, until the cessation of such work, about three years after starting. They showed 53 to 117 nCi/ml urine, or about two to five times the maximum permissible concentrations. Figure 3 and Table 4 show the values. A

Figure 3

Tritium in Urine
Case B.2



very rough estimate of the whole body dose is given by Minder [2] as 1000 to 2000 rem within a period of three years. This estimated whole body dose is, however, extremely uncertain as no measurements are available for most of the working period, and the extrapolation from the period after B.2 had stopped work with tritium may be incorrect by an order of magnitude. More probably the "body water dose" lies between a few hundred and a maximum of one thousand rem.

The case of B.2 is most informative, as many measurements are available for the period after he had stopped working with tritium. After an initial decrease in the tritium concentration in the urine with a biological half-life of about 20 days, an increase and a second period of excretion were observed. This second period can, following the above model by Gottschewski [5], be described by two exponential functions with half-lives of 10 and 139 days, respectively, the complete excretion formula being:

$$A(t) = 24.9 \times e^{-0.068t} + 11.9 \times e^{-0.005t}$$

(For "t" = 0 the 140th day after cessation of work with tritium was taken).

In another interpretation of the results given by Minder [2] and using the day on which work was stopped as "t" = 0, two exponential functions appear with 30 and 230 days half-life, respectively. The formula thus becomes:

$$A(t) = 200 \times e^{-0.022t} + \times e^{-0.003t}$$

to which the body water component with 10 to 12 days must be added. Other interpretations are, of course, also possible.

In Figure 3 the T = 139 days (following Gottschewski's model), T = 227 days (representing the second exponential function given by Minder), T = 168 days (which graphically fits best to the last part of the excretion data), and T = 90 days and T = 257 days (corresponding to the biological half-life quoted by ICRP for nitrogen and phosphorus, respectively) are shown for optical comparison. At least one long-term component of tritium has, for this case as well as others reported in the literature, half-life in between the values for nitrogen and phosphorus, similarly also sulphur and iodine, whereas other main elements in a body, i.e., hydrogen, carbon, oxygen, and sodium, show half-lives between 10 and 14 days.

Might there be a connection of the metabolism of the former elements, particularly N or/and P, with that of tritium, and can we assume in this case a microscopically uniform distribution of those elements within the cells and also the intercellular substances? Should one think of the possibility of some tritium being specifically bound to the DNA, not uniformally distributed throughout the cell? How would this affect the microscopic dose distribution and damage to the DNA by radiation of isotopic effect?

After working exactly three years with tritium, B.2 became so seriously affected that he stopped working with radioactive substances. Fifteen and a half months later he died, with the symptoms of Panmyelocytopenia. Some of the blood counts are included in Table 2, unfortunately covering only the period before stopping work.

In Table 3 the concentrations of tritium in body tissues after death are given. They were around 6 to 12 times as high as in the body water (urine). It might be interesting to know that an elution of the tissue water with formalin even over months did not result in an elution of tritium; this was obviously strongly bound to organic compounds. The specific concentrations in the organic compounds, subtracting the water content from the tissue mass, could therefore range even between 22 and over 40 times the concentration of tritium in urine. An evaluation of the body burden based on urine concentrations would in this case have led to a serious underestimation.

In the control of workers using tritium, one can therefore never rely on a single measurement of tritium in urine. One must measure the excretion function and assume the existence of organically bound tritium in reasonable quantities in case the half-life of tritium is longer than 15 to 20 days. An experiment on human volunteers with a single known intake of tritium and maximum possible extremes for the water turnover (one volunteer drank as much as he could, four to five liters per day; another drank as little as possible), showed half-lives between 5.5 and 19 days for a period of 60 to 90 days; then the function seemed to change. Unfortunately, it was not possible to follow up the excretion for a longer time, as amounts became too low for significant results to be obtained with the method of measurement used.

Conclusions

We may draw a number of conclusions, or at least be stimulated to carry out further research, from these cases:

1. There is strong evidence that two people died and several others were seriously affected by the incorporation of tritium in the course of the production of tritiated luminous compounds.

2. These cases of chronic intake of tritium confirm the results obtained in other cases of accidental tritium incorporation. A part of the tritium is transferred to organic compounds and the biological half-life of tritium in the organic compartment or compartments may vary between 30 and 230 days or more. Nitrogen, sulphur, and phosphorus all have half-lives in this range and this may give some indication where tritium bound to organic material may be located.

3. When sufficient time has elapsed after the last intake of considerable activities, then the concentration of tritium in the body can be higher than that measured in urine samples. This may lead to a serious underestimation of the actual body burden. A single measurement is therefore not significant in cases where the possibility of a tritium incorporation, two months or earlier before the measurement, cannot be excluded. Several measurements have to be performed to evaluate the function for the excretion in this case.

4. There is some evidence that one to two percent of the total incorporated activity is transferred into organic compounds and, in the case of chronic incorporation, about 12% (following a given model: the figure may lie between 10 and 20%) of the actual tritium content of the body is bound to organic material. If a microscopically uniform distribution of tritium is assumed for both the body water and the organic compounds in which tritium is incorporated, then about 85% of the integrated absorbed dose comes from tritium in the body water and about 7 to 15% from tritium in organic material (following the mentioned model). The question remains, however, as to whether

this assumption is correct. If, for example, tritium participates preferentially in anabolism rather than catabolism and is incorporated into the DNA (the similar half-lives of nitrogen, etc., could indicate this), then a somewhat nonuniform distribution might result and the damage to the cell nucleus or to the mitochondria might be much higher than would be expected on the assumption of uniform distribution. In this case, tritium transferred to the organic phase might be as important, or even more important, than that in the body water.

5. The whole body dose, calculated conventionally for the two cases in which death occurred was probably about 300 rem within six years for one and less than 1000 rem within three years for the other. The biological effect in both cases was an almost complete destruction of the red bone marrow, indicated clinically by a slow but continuously progressive hyperplastic anemia rather than by changes in the white cell count. A second person involved in Case A, who received an estimated dose of about 150 rem over a period of nearly four years, also showed a slight hyperplastic anemia but no severe damage to health. The higher evidences of changes in the red cell blood counts, compared with no significant changes in the white blood cells, in case of chronic incorporation of tritium has to be stressed.

6. The dose in Case A, which amounted to about 10 times the maximum permissible dose (MPD) recommended by ICRP, and which is in the range of "Tolerance doses" established 35 years ago, possibly may not be considered sufficient to cause even subacute illness. The possibility may have to be considered that the conventional method of determining the dose commitment for tritium may be unsuitable for assessing the biological effect. This may be due either to a nonuniform distribution of tritium in the organic phase or to an isotopic effect of tritium. The MPD may not be so unrealistic and conservative as many people think today.

If the conclusion of Bond is correct, that the maximum permissible values for tritium recommended by ICRP are too conservative by a factor of five to eight and the other considerations mentioned above are not relevant, then the conclusion would be that the value for the maximum permissible dose is very near to a hazardous radiation exposure.

7. Two cases of death have been presented and theoretical evaluations and a number of hypotheses given; the result is a number of question marks.

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CHROMOSOMAL CHANGES IN CHINESE HAMSTER CELLS FOLLOWING CUTANEOUS EXPOSURE TO TRITIATED LUMINOUS COMPOUNDS

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Early theoretical calculations suggested that tritiated thymidine (^{3}HTdR) should be considered more toxic to mammalian systems (by several orders of magnitude) than tritiated water [1,2]. Subsequent experiments demonstrated that cytological damage caused by ^{3}HTdR included chromosomal abnormalities with levels of 1 μCi ^{3}H/g of body mass [3,4]. On the basis of tritium-induced chromosome damage observed by others, this study investigating the chromosome damage to Chinese hamsters resulting from percutaneous absorption of tritium activity from luminous compounds was undertaken.

Methods and Materials

Luminous compounds from two different companies were used in this study. The compound from Radium Chemical Company (RH) had a specific activity of 20 mCi/g, and the compound from 3M Company (MH) had 200 mCi/g specific activity. Each hamster was exposed to 5 mg of luminous compound; thus the approximate activities applied were 100 μCi to the RH group of hamsters and 1000 μCi to the MH group.

Prior to the experiment, 30-day-old Chinese hamsters were placed in metabolic cages to become acclimated. One day pre-exposure the hair was clipped from the animal's back, with care being taken not to break the skin. The following morning 5 mg of the luminous compound was weighed onto an area 1 cm^2 on a piece of plastic wrap mounted on surgical tape. The animal was placed upon the bandage with the compound facing the clipped area and the bandage carefully taped in place. An additional wider piece of tape was fastened around the animal's body to secure the inner tape and prevent loss of luminous compound.

Eight hours later the bandages were removed, and the remaining compound washed off carefully to avoid its spread over a larger area. Urine samples were collected every 24 hours and aliquots counted in a liquid scintillation counter to monitor for tritium excretion. The results were recorded as nCi/ml of urine. Control animals were handled in the same manner, but received no compound.

Groups of hamsters were sacrificed for tissue culture at 7 and 30 days post exposure. Lung and kidney tissues were grown in Leighton tubes for chromosome studies. Replicates of each exposure period were run. The cells were allowed to grow for 72 hours, then Colcemide was added and allowed to act for three to four hours, followed by hypotonic treatment, fixation, and staining.

The stained slides were scanned under low magnification for metaphase cells, these being the most suitable for chromosome analysis. Metaphase cells, spread and fixed in a manner to permit clear interpretation, were then examined under phase oil immersion and classified as aberrant or nonaberrant.

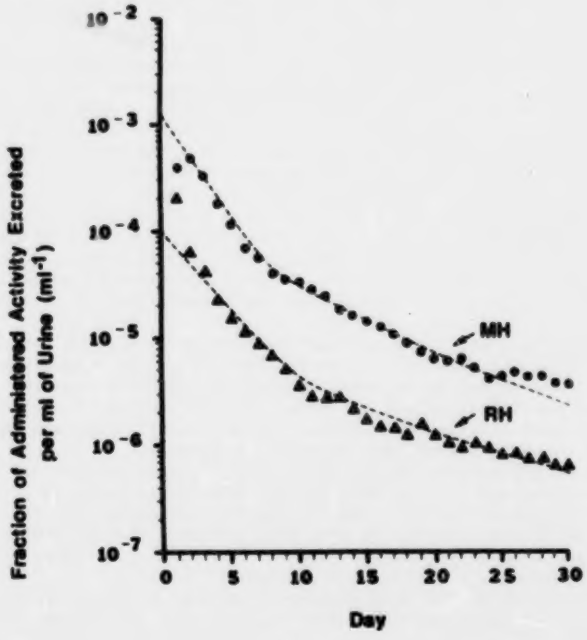
Results and Discussion

A cell was considered to be nonaberrant if there was no evidence of chromosomal change (aneuploid cells were not considered aberrant). Metaphase cells containing chromosomes with structural abnormalities were counted as aberrant. These changes were classified as chromatid and isolocus breaks, dicentrics, rings and exchanges. Chromosome or chromatid gaps were not scored as abnormalities, nor were fragments counted as chromosomes. Endoreduplications, which occurred in 0.23% of total cells scored, were classified as aberrant.

The majority of the aberrations were chromatid breaks with the frequency of other aberrations occurring in the following order: isolocus breaks, dicentrics, exchanges, endoreduplications and rings. This order held true for both compounds and for both exposure periods. There was some increase in the number of isolocus breaks at 30 days, but these did not outnumber the chromatid breaks. The results indi-

Figure 1

Chinese Hamster Skin Absorption of Tritium Activity from Luminous Compounds



cate most of the damage occurred during the G2 phase of the cell cycle. The slight increase in the frequency of isolocus breaks at 30 days could be due to second generation cells or to damage during the G1 phase of the cell cycle.

Fifteen and sixteen animals were exposed to RH and MH compounds, respectively. Figure 1 shows the daily urinary radioactivity following cutaneous absorption of the two compounds divided by the administered activity. The curves indicate the relative absorption of the tritium activity from the two compounds. Levels of tritium in urine collected from control animals were on the order of 0.01 nCi/ml.

The lung and kidney chromosome damage, as percent of aberrant cells, from the MH compound ranged from 5.4% at seven days to 7.1% at 30 days. The RH compound produced approximately the same percent of aberrations at both time periods (Table 1). As evidenced from Figure 1, more tritium activity was absorbed from the MH compound than from the RH compound. One might expect to find a higher aberration percentage associated with the MH compound. This was not the case. Possibly the tritium from the RH compound was contained in a form that produced more damage. This aspect could not be investigated further.

Table 1

Effect of Two Tritiated Luminous Compounds on the Percent of Aberrant Cells in Adult Hamster 7 and 30 Days After Exposure

Compound and Administered Activity	Tissue	7 days			30 days		
		Total Cells Scored	Aberrant	Percent Aberrant ±2σ	Total Cells Scored	Aberrant	Percent Aberrant ±2σ
MH - 993 μCi per hamster	Kidney	408	22	5.4 ± 2.2	406	25	6.2 ± 2.4
	Lung	643	39	6.1 ± 1.9	987	70	7.1 ± 1.6
RH - 95 μCi per hamster	Kidney	975	75	7.7 ± 1.7	421	30	7.1 ± 2.5
	Lung	466	33	7.1 ± 2.4	624	42	6.7 ± 2.0
Controls	Kidney	761	7	0.9 ± 0.7	918	4	0.4 ± 0.4
	Lung	501	4	0.8 ± 0.8	1297	17	1.3 ± 0.6

The standard deviation (σ) of the percent of aberrant cells was calculated by assuming a binomial distribution. Thus,

$$\sigma = \sqrt{\frac{P(1-P)}{N}}$$

where P is the fraction of aberrant cells and N is the total number of cells scored. The error presented in Table 1 is ±2σ. From these data, it is apparent that there is a significant difference between the test animals and the controls.

Conclusions

The present study has investigated the possibility of chromosomal damage following skin absorption of tritium from luminous compounds in the Chinese hamster. The following conclusions were reached:

1. Tritium was absorbed through the skin of Chinese hamsters as indicated by the appearance of significant amounts of tritium in the urine of the test animals. There was only a slight indication of tritium in the urine of the control animals.

2. Chromosomal aberrations were produced at 7 and 30 days in both lung and kidney tissues. With both compounds tested, the percentage of lung and kidney cells containing aberrations was several times greater than the control values.

3. The production of chromosome aberrations is attributed to the luminous compound (tritium compound plus fluor) since the comparison of controls and test animals was a comparison between animals exposed to luminous compounds and those exposed to no compound.

4. The types of aberrations found most often were chromatid breaks, which demonstrated that the most damage occurred during the G2 phase of the cell cycle.

ACKNOWLEDGMENTS

Work was initiated while this Laboratory was a component of the Bureau of Radiological Health, U.S. Department of Health, Education and Welfare.

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INFLUENCE OF IRRADIATION ON TRITIUM INCORPORATION IN HAIR

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In a recent work [1] it was observed that the incorporation of tritium as tritiated water into the hair of rabbits is influenced by the amount of nuclide given subcutaneously. The incorporation in rabbits given 38 mCi per animal was three times higher than in rabbits given 10 and 100 times lower doses. This effect has also been observed with other nuclides, usually finding the depression of incorporation at higher dose levels. It is suggested that this is probably caused by radiation effects in the skin and hair matrix [2,3].

This paper attempts to find if the effects observed in rabbits can be repeated in other mammals and to determine the effect of external irradiation on the tritium incorporated into the hair. Also, a similar experiment with tritium was carried out on rats. The results obtained are compared with those found in experiments with other radionuclides.

Materials and Methods

The hair of Wistar inbred rats was plucked out from an area of about 40 cm^2 on the dorsal part of the trunk. New anagen hair emerged above the skin on the 11th day after plucking. On the 15th day, the rats were divided into eight groups of 12 animals each, and were placed in metabolic cages to prevent external contamination with excreta. On the same day, these rats, weighing approximately 200 g, were given tritium subcutaneously in single doses of 4.5, 45, 450, and 2265 μCi per animal in the form of tritiated water in 0.9 percent sodium chloride aqueous solution containing 0.5 percent of phenole.

Total body irradiation with X-rays of two groups of animals was performed on the same day before administration of 4.5 μCi of tritiated water per animal. Two groups of rats were irradiated with 50 and 200 rads of X-rays (250 kV, 15 mA, 1.5 mm Cu + 1.0 mm Al filtration, 27.1 R per min). The homogeneity of irradiation was measured with thermoluminescent dosimeters.

On the 21st day after tritium administration, the anagen hair was plucked from the same area. Samples of 100 mg of hair were combusted in two liter flasks filled with oxygen and the resulting water was mixed with liquid scintillator. The tritium activity was measured in a Packard Tri-Carb Liquid Scintillation Spectrometer, Type 3315. The statistical counting error at the confidence level of 0.95 did not exceed five percent, and the chemical yield of the procedure was 93 percent. Incorporation values are expressed in percent of administered dose per gram of hair.

Results and Discussion

The incorporation of tritium varied from 0.027 to 0.042 percent of injected dose per gram of hair, Table 1. These results are about ten times higher than values found in a previous experiment with rabbits. The higher values in rats may have been caused by species differences.

Table 1

Incorporation of Tritium in Anagen Hair of Rats in Percent of Injected Dose Per Gram of Hair

Tritiated Water µCi/animal	Radiation Dose (rems)	Percent Incorporated ± Standard Deviation
4.5	0.075	0.042±0.007
45.0	0.75	0.031±0.007
450.0	7.5	0.030±0.004
2265.0	35.0	0.027±0.004

The tritium content of telogen hair in rabbits and rats was about one order of magnitude lower than in anagen hair. This indicates that licking and other causes of external contamination are not responsible for the observed difference between the two species. This difference cannot be explained as a simple effect of the rate of isotopic dilution of tritium in the fleece, as the mass of hair per skin surface unit is similar in both animals. Also, the ratio of hair mass to body weight in rabbits is about 2.5 times lower than in rats, which would suggest that, with the same tritium doses per gram of body weight, the incorporation values in rabbits should be correspondingly higher than those in rats, Table 2. The inverse effect found in the experiments suggests that metabolic differences between the two species are responsible for the observed phenomenon.

Table 2

Comparison of Hair, Skin and Body Weight Proportions Between Rat and Rabbit

	Rabbit (4400 g)	Rat (200 g)
hair mass/skin surface, g/cm^2	0.025	0.020
hair mass/body weight, g/kg	11.0	27.6
skin surface/body weight, cm^2/g	0.45	1.38

The internal radiation doses received by rats in this experiment, calculated by the method of Lambert and Clifton [4], ranged from 0.075 to 35.0 rem depending on administered doses of tritiated water. In this dose range, a decrease of incorporation values was observed with increasing tritium doses, i.e., an effect opposite to that found in rabbits. Statistical analysis performed by Student's t test indicates that the differences observed between particular groups of rats are statistically significant at the 0.99 confidence level.

To find the possible effect of external irradiation on incorporation, the animals were irradiated with 50 and 200 rad of X-rays. The results, presented in Table 3, indicate that irradiation of rats slightly increases the tritium incorporation in hair as compared with nonirradiated controls. The differences observed between controls and 50 and 200 rad groups are statistically significant at 0.98 and 0.90 confidence levels, respectively. However, lower incorporation values were observed at the external doses of 200 rad than at 50 rad, the difference being statistically significant at the 0.80 confidence level.

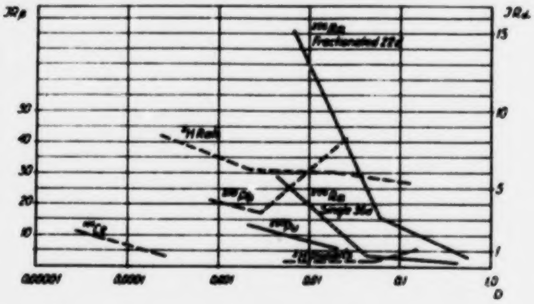
Table 3

Effects of External Irradiation
On Incorporation Rate of Tritium in Hair
in Rats Given 45 μCi Tritiated Water Per Animal

Radiation Dose rad/animal	Percent Incorporated ± Standard Deviation
50	0.035±0.007
200	0.032±0.003
controls	0.029±0.005

Figure 1

Incorporation Rates JR, in Hair of Rabbits (all nuclides) and Rats (tritium)
JR = % Administered Dose Per Gram of Hair X 10^{-3}
D = Arbitrary Units of Internal Irradiation (μCi/g X MeV)



In Figure 1, a comparison of internal irradiation of animals in experiments with tritium and other radionuclides was made by normalizing the injected doses of nuclides into arbitrary D units, i.e., μCi of administered nuclide per gram of body weight multiplied by effective energy (in MeV) of the nuclide. The values of effective energies for nuclides are from reference 5. As may be seen from this Figure, the tritium curve corresponding to the changes in incorporation with the level of administered dose is surprisingly flat as compared with the curves found in experiments with other radionuclides.

To elucidate the causes of the observed phenomenon, a more detailed investigation is needed with studies of a wider range of both tritium and external radiation doses.

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CHAPTER FIVE
Kinetics of Tritium in Biological Systems

KINETIC AND EQUILIBRIUM ISOTOPE EFFECTS OF TRITIUM SUBSTITUTION

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The principles governing the effects of isotopic substitution on the rate constants and equilibrium constants of chemical reactions have been understood for some time [1]. The fact that molecules differing only in the atomic weight of one of the constituent atoms do indeed have slightly different chemical properties has important applications in the study of reaction mechanisms, intramolecular and intermolecular forces, etc. Practically, isotopic fractionation on an industrial scale produces heavy water for use as a moderator in nuclear reactors.

These same general principles will apply to molecules containing tritium, and if the labeling is only at the tracer level, normal isotope effects will be more important than the effects of tritium β-radiation, but not necessarily more important than chemical effects resulting from the transmutation of tritium nuclei. However, in reviewing the literature for examples of isotope effects due to tritium substitution, one notes a conspicuous lack of data compared with those available for the other rare hydrogen isotope, deuterium.

Isotopic Fractionation of Tritium in Chemical Equilibria

Statistical mechanics provides a link between molecular properties and thermodynamic quantities. Thus it is possible to relate the equilibrium constant for a chemical reaction to the partition functions of the reactants and products. These, in turn, are calculable from vibrational frequencies, moments of inertia, and molecular weights. Strictly speaking, this is correct only for ideal gases.

Consider a special type of equilibrium in which tritium and protium are exchanged between two different chemical species:

$$AH + BT \rightleftharpoons AT + BH$$

The equilibrium constant is

$$K = \frac{[AT]\,[BH]}{[AH]\,[BT]} = \frac{Q_{AT}\,Q_{BH}}{Q_{AH}\,Q_{BT}} \qquad (1)$$

where the Q's are partition functions. Bigeleisen and Mayer [2] have shown that the partition function ratio differs from unity only because of quantum mechanical contributions from vibrational partition functions. Thus,

$$K = f_{AT}/f_{BT},$$

where $$f = \prod_{i=1}^{3N-6} \frac{u_{2i}\,(1 - e^{-u_{1i}})}{u_{1i}\,(1 - e^{-u_{2i}})} \exp[(u_{1i} - u_{2i})/2] \qquad (2)$$

In this expression, N is the number of atoms in the molecule, and subscripts 1 and 2 refer to the protium- and tritium-containing molecules. In terms of vibrational frequency ν, Planck's constant h, the Boltzmann constant k_B, and the absolute temperature T, the dimensionless quantity u is given by

$$u = h\nu/k_B T.$$

In the preceding expression for f, classical statistical factors not contributing to isotope fractionation are omitted.

If the necessary vibrational frequencies are known, or can be computed from vibrational force constants, then f of Eq. 2 can be evaluated. Even if this is not practicable, the expression provides some insight into the basis for fractionation of tritium between two molecules:

1. Force constants of the two molecules must differ at the position of isotopic substitution.
2. The heavy isotope (in this case, T) will concentrate in the molecule in which it is most strongly bonded.
3. The major part of the effect, at "reasonable" temperatures, results from the last term of Eq. 2, the difference in zero-point vibrational energies. This is shown schematically in Figure 1.

Figure 1

Potential energy of hypothetical diatomic molecules containing protium or tritium as a function of internuclear distance. The zero-point vibrational levels are indicated by horizontal lines. Energy minimization will favor the accumulation of tritium in the reactant and protium in the product.

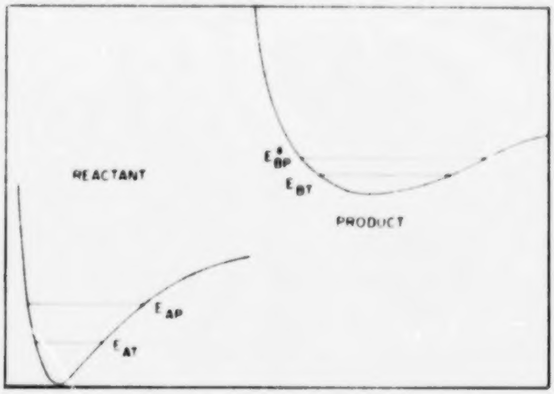


Table 1

Equilibrium Fractionation of Tritium Between Water (Liquid) and Hydrides at 25°C

Compound	α_T	Method	Reference
H_2	6.85	Experimental	23
HF	1.27	Vib. Frequencies	24
HCl	3.37	Vib. Frequencies	24
HBr	4.50	Vib. Frequencies	24
HI	6.79	Vib. Frequencies	24
H_2S	3.30	Exptl. D/H + Vib. Freq.	25
H_2Se	4.20	Exptl. D/H + Vib. Freq.	25
NH_3	1.06	Experimental	26
PH_3	2.45	Exptl. D/H + Vib. Freq.	27
AsH_3	2.84	Exptl. D/H + Vib. Freq.	28

4. The value of k approaches unity at high temperatures.

Since the major concern is with T in the form of HTO, several examples of equilibria involving this molecular species are given in Tables 1 and 2. The data are given in the form of the fractionation factor: $\alpha_T \equiv (T/H)\ \text{water}/(T/H)\ \text{compound}$.

In Table 1, note that α_T increases systematically towards the bottom of the periodic table in the hydrogen halide series, as the vibrational force constant becomes smaller than those of H_2O. A similar effect is found for the hydrides of Groups V and VI.

In Table 2 data are collected for groups which are typical of those important biologically, and which (with the exception of -CH) can exchange rapidly with water. The only large fractionation of tritium is for the -SH group, and here T will concentrate in the water. It is somewhat surprising that α for the -OH groups of cellulose is so different from that of small molecules containing the -OH group. A similar comment applies to the difference between the peptide -NH groups of polyalanine and the amide group of formamide. If these differences are real, they must result from the additional intramolecular forces in the large polymer molecules.

A word about the sources of data for these tables is in order. As was mentioned earlier, relatively few experimental data for T-containing compounds can be found in the literature. For diatomic molecules such as the hydrogen halides, the vibrational frequencies of all isotopic species can be calculated exactly if the frequency for one species is known. This cannot be done for polyatomic molecules,

Table 2

Equilibrium Fractionation of Tritium between Water (Liquid) and Various Compounds at 25°C

	α_T	Method	Reference
-OH groups			
H_2O (vapor)	1.094	Experiment	29
CH_3COOH (aq)	1.00	Experiment	30
CH_3OH (liquid)	1.02	Exptl. D/H + Vib. Freq.	31
Cellulose (solid)	0.7	Experiment	32
-NH groups			
NH_3	1.06	Experiment	26
$HCONH_2$	1.03	Exptl. D/H + Vib. Freq.	33
Ribonuclease (aq)	1.0	Experiment	34
DL-polyalanine (aq)	0.83	Experiment	35
-SH groups			
H_2S	3.30	Exptl. D/H + Vib. Freq.	25
CH_3SH (liquid)	3.31	Exptl. D/H + Vib. Freq.	36
-CH groups			
$HC{\equiv}CCH_3$	1.28	Exptl. D/H + Vib. Freq.	37
$HC{\equiv}CCH_3$	1.42	Vib. Freq.	38
CH_3SH	1.57	Vib. Freq.	38
C_6H_6	1.17	Estd. from D	39

The exchanging hydrogen atoms are in bold-face type.

where a number of force constants are required.

Bigeleisen [3] has proposed a method for relating α_T for tritium-protium exchange to the analogous fractionation factor for deuterium-protium exchange: $\alpha_D \equiv (D/H)water/(D/H)compound$. At very high temperatures, the relation

$$\frac{\ln \alpha_T}{\ln \alpha_D} = \frac{4}{3}$$

is rigorously correct. At other temperatures, it becomes necessary to use Eqs. 1 and 2, with the relations

$$\alpha_T = f_{AT}/f_{BT} \qquad \text{and} \qquad \alpha_D = f_{AD}/f_{BD} .$$

If two constants a and b are defined by

$$\ln f_{AT} = a \ln f_{AD} \qquad \text{and} \qquad \ln f_{BT} = b \ln f_{BD}$$

then

$$\frac{\ln \alpha_T}{\ln \alpha_D} = \frac{a \ln f_{AD} - b \ln f_{BD}}{\ln f_{AD} - \ln f_{BD}}$$

$$= a + \frac{(a - b) \ln f_{BD}}{\ln \alpha_D} . \tag{3}$$

For the cases labeled "Exptl. D/H + vib. freq." in Tables 1 and 2, the vibrational frequencies, f_{AD}, and f_{AT} have been calculated. At 25°C the quantities a and b are always close to 1.4 for these examples. Hence (a - b) is small, and the second term of Eq. 3 is usually small. However, the actual values in Tables 1 and 2 were obtained from experimental determinations of α_D and the use of Eq. 3 with calculated values of a and b. This should be more precise than the use of the general expression suggested by Bigeleisen [3]

$$\ln \alpha_T/\ln \alpha_D = 1.33 \text{ to } 1.55 .$$

In particular, if α_D is close to unity, the second term in Eq. 3 can become large. Of course, in that case, the isotopic fractionation is small and there is little practical interest in the equilibrium.

Stern and Vogel [4] have calculated the ratio $\ln\alpha_T/\ln\alpha_D$ over a wide temperature range for many equilibria, and find numerous anomalies where the relative values of α_D and α_T are outside the expected range of 1.33-1.55.

To summarize the data of Tables 1 and 2, it seems unlikely, in the opinion of the author, that equilibrium fractionation of tritium from water in biological systems would lead to significant concentrations in other molecules within the biological system.

The Effect of Tritium Substitution on Reaction Rate Constants

Unfortunately, the relationship between molecular properties and equilibrium constants (via statistical mechanics) does not have an analogy applicable to rate constants. Only a few rate constants have been calculated a priori, and these are for systems of a few atoms. A simplification that has often been used is the activated complex theory (ACT) [5], in which the "activated complex" is assumed to be in equilibrium with reactants. This theory leads to the formulation of a rate constant for a hypothetical reaction

$$AH + B \longrightarrow A + HB$$

in the form

$$k = \frac{k_B T}{h} \; \frac{Q^{\ddagger}}{Q_{AH} \; Q_B} \qquad (4)$$

The term $Q^{\ddagger}$ is the partition function for the "activated complex," a particular configuration of the reaction system at the top of the potential energy barrier that separates reactants from products. The ratio of two rate constants for reactants differing only isotopically can be expressed conveniently by ACT [6]. Thus, for the reactions

$$AH + B \longrightarrow A + HB$$

and

$$AT + B \longrightarrow A + TB$$

we have

$$\frac{k_H}{k_T} = \frac{\nu_T^{\ddagger} \, f^{\ddagger}}{\nu_P^{\ddagger} \, f_{AT}} \qquad (5)$$

The f's in this expression have the same form as those of Eq. 2, except that $f^{\ddagger}$ contains one less term, that corresponding to motion along the reaction coordinate. This motion contributes only the ratio $\nu_H^{\ddagger}/\nu_T^{\ddagger}$ because it does not correspond to a bound vibrational state; neither a vibrational partition function nor a zero-point energy term is present.

The important things to note about Eq. 5 are:

1. As in the case of the equilibrium fractionation of isotopes, only vibrational properties of the reactants and the activated complex are important.

2. For the ratio k_H/k_T to differ from its high-temperature classical limit of $\nu_H^{\ddagger}/\nu_T^{\ddagger}$, vibrational force constants at the position of isotopic substitution must change between the reactant and the activated complex. Usually, force constants are weaker for the activated complex, and therefore the heavy isotope concentrates in the reactant instead of the activated complex. This leads to a "normal" isotope effect in which the light molecule reacts more rapidly than the heavy one.

3. Thus, kinetic isotope effects provide a probe for the force constants of the activated complex if those of the reactant are known. These force constants are determined, in turn, by the potential energy surface in the immediate vicinity of the activated complex configuration.

4. In reactions where hydrogen atoms or ions are being transferred, it is often necessary to take into account the wave nature of the hydrogen particle which enables it to "tunnel" through the potential energy barrier even when it does not possess enough energy to get over the top [7]. This leads to another term in Eq. 5 which can be calculated if the shape of the potential barrier is known.

The application of kinetic isotope effects to chemical kinetics fall roughly into two categories:

1. Qualitative investigations of reaction mechanisms, and

2. Investigations of various methods of calculating potential energy surfaces.

One of the earliest applications of a tritium isotope effect to a mechanistic study is the classic work of Melander [8] on aromatic (Ar) electrophilic substitutions. In the reaction scheme

$$ArH + NO_2^+ \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} ArHNO_2^+ \xrightarrow{k_2} ArNO_2 + H^+$$

ordinary kinetic experiments do not distinquish between a mechanism involving the formation of $ArHNO_2^+$ as an intermediate, and a direct displacement of a proton by the nitronium ion.

Melander measured relative rates of reaction of aromatic compounds and their tritiated analogs. As Table 3 indicates, the isotope effects are rather small. A reasonable interpretation of these results is that the direct mechanism is not followed, and that in the complex mechanism, step (1) is the rate-determining step.

Table 3

Kinetic Isotope Effects in the Nitration of Aromatic Compounds (from Melander [8])

Compound	k_H/k_T at 77°C
Benzene-T Nitrobenzene-T	1.19 - 1.35
Toluene-2-T p-Nitrotoluene-2-T	1.18 - 1.92
Bromobenzene-2-T p-Nitrobromobenzene-2-T	1.23 - 1.28
Naphthalene-α-T	1.30

Nitration continues after the first substitution to give a mixture of reaction products indicated by the brackets.

Kinetic isotope effects have been used to test various detailed models for activated complexes. Because of the difficulty of applying rigorous quantum mechanical methods to such problems, semi-empirical and empirical techniques have been developed for constructing potential energy surfaces and obtaining force constants [9].

A completely empirical approach, the bond energy-bond order (BEBO) method of Johnston and Parr [10] has proven to be very useful in predicting activation energies and isotope effects in hydrogen abstraction reactions. The reaction system, to a first approximation, can be considered to be a three-atom system

$$AH + B \longrightarrow A + HB$$

The BEBO method has been used to calculate kinetic isotope effects of tritium substitution in several reactions of this type.

Figure 2

Rate constant ratios for reactions of the type $R + P_2/PT \rightarrow RP/RT$ as a function of T^{-1}. Bond dissociation energies (D_e) of reactant and products are tabulated. O, Cl + P_2/PT, data from Ref. 44; □, Br + P_2/PT, data from Ref. 45.

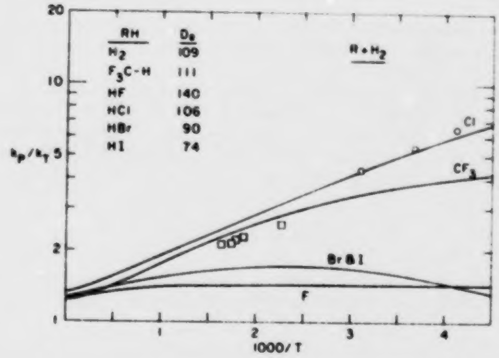


Figure 3

A plot similar to Figure 2, but for reactions of halogen atoms with CP_4 and CP_3T.

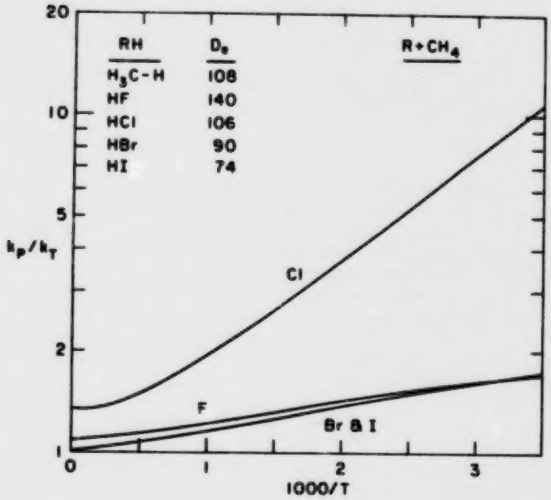
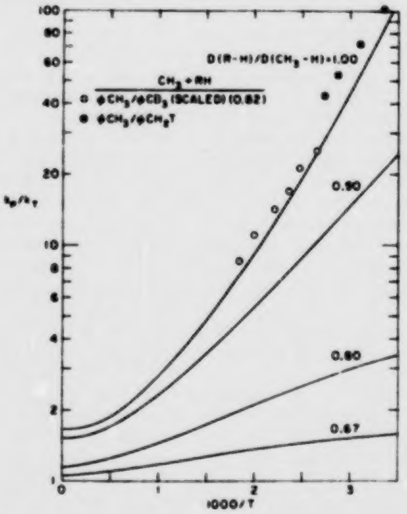


Figure 4

A similar plot for reactions of the type $CH_3 + RP/RT \rightarrow CH_3P/CH_3T$. The different lines are for different ratios of bond dissociation energy $D_e(R - H)/D_e(H_3C - H)$. ●, $C_6H_5CP_3/C_6H_5CP_2T$ (Ref. 15); O, from data for $C_6H_5CP_3/C_6H_5CP_2D$ (Ref. 16), calculated with the assumption that $\log(k_P/k_T) = 1.465 \log(k_P/k_D)$.



The results are shown in Figures 2 to 4, where the rate constant ratio k_H/k_T is plotted against T^{-1}. Because the BEBO formulation relates properties of the activated complex to properties of the reactants and products, one might expect a correlation between the quantity k_H/k_T and the bond dissociation energies D(A-H) and D(B-H). Calculated values of k_H/k_T do indeed exhibit a trend. For example, in the reactions of radicals and atoms with H_2 and HT, Figure 2, the maximum isotope effect is predicted for the reactants Cl and CF_3. The bond dissociation energies (De) for HCl, CF_3H, and H_2 are 106, 111, and 109 kcal/mole. In the activated complexes for these two reactions, the transferring hydrogen atom is partially bonded to both the attacking group and the remaining hydrogen atom. The vibrational frequencies of the activated complex are lower than that of H_2 and a large isotope effect results. By contrast, the activated complex for a very endothermic reaction such as H_2 + Br closely resembles the product molecule, HBr; conversely, in an exothermic reaction such as H_2 + F, the activated complex is a nearly unperturbed H_2 molecule with a very weak bond to F. In either case, the vibrational terms in f^* tend to cancel those in f_{H2}, and small isotope effects are predicted.

A similar type of correlation was predicted in a qualitative way by Hammond [11], and it is interesting to see it here in a more quantitative form. Similar results have also been obtained by Johnston [12] using both the BEBO method and a different method for calculating potential energy surfaces.

Unfortunately, this predicted correlation is not found experimentally, as the data in Figure 2 for reactions of H_2 with Br and Cl indicate. This is even more striking in the case of competition between H_2 and HD, where more experimental information is available. Here the points for Cl, CF_3, Br, and I are very nearly on a single straight line.

Figure 3 shows analogous calculated results for hydrogen abstraction by halogen atoms from methane. No experiments have been done with tritium-labeled methane, but in the reactions Cl + CH_2D_2 and Cl + CH_4/CD_4 it is found that the observed k_H/k_D values are in good agreement with those predicted [13]. However, in the reaction Br + CH_4/CD_4 [14] the k_H/k_D values are in better agreement with those calculated for Cl + CH_2D_2 than for Br + CH_2D_2. In other words, the experimental results agree with the *maximum* predicted isotope effects.

Figure 4 shows the predicted trend in isotope effects for reactions in which the bond dissociation energy of the C-H bond being broken (rather than the energy of the bond being formed) is changed. Again, the maximum isotope effect is predicted for the symmetrical activated complex which results from equal energies of the bonds being formed and broken. The only available data, for competitive H and T abstraction from $C_6H_5CH_2T$ by methyl radicals [15], again agrees with the *maximum* predicted effect, rather than with the effect predicted for the toluene molecule with D(C-H), about 80% that of D(C-H) in methane. Also shown on this plot are data for the analogous deuterium isotope effect [16] which have been "scaled up" by using an expression similar to Eq. 3.

What is wrong with the predicted isotope effects in these systems? One possibility, of course, is that the activated complex properties calculated by the BEBO method are not generally correct. The test of this will only come with good *ab initio* quantum mechanical calculations of potential energy surfaces for these systems. This goal is just now coming within the reach of quantum chemists, and depends to a considerable extent on the availability of enormous amounts of computer time.

A second possibility, which is suggested by recent work of Morokuma and Karplus [17], is that the activated complex theory leading to Eq. 5 is, in fact, invalid for certain systems. Their work (although limited to a few cases) indicates that ACT breaks down for very "unsymmetrical" reactions, i.e., those that are highly exothermic or endothermic. This would fit in with the pattern of the results reported here.

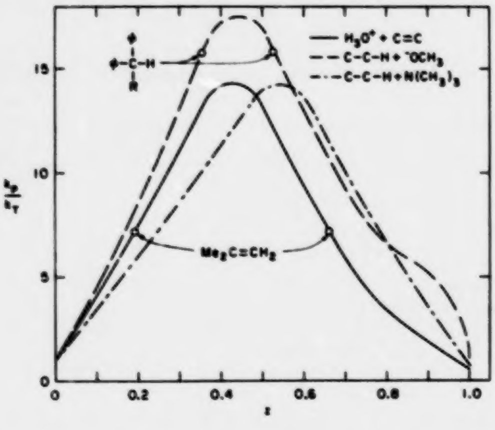
The group of hydrogen abstraction reactions just discussed is of considerable interest to chemical kineticists, particularly those who specialize in gas-phase reactions. However, they are rather far removed from the types of reactions one might expect to be important in biological systems at room temperature or thereabouts. There is a large class of reactions which are important in biochemistry and in which large hydrogen isotope effects are found: proton-transfer reactions. A large number of these reactions in the category of acid or base catalysis have been investigated. Again, most of the isotope effect experiments have utilized deuterium, frequently by comparing reactions in D_2O and H_2O. Since the solvent properties of these two species are significantly different, the interpretation of such experiments is complicated, and in this respect, the use of tritium at the tracer level offers certain advantages.

The theoretical treatment of proton-transfer reactions, with few exceptions, has been even more empirical than the treatment of hydrogen abstraction reactions. Typically, vibrational force constants for a simplified model of the activated complex are related by some arbitrary parameter to those of the reactant and product. Then a range of values for k_H/k_T is predicted, within the allowed range of the parameter. For example, More O'Ferrall and Kouba [18] use as a model for base catalysis the reaction system

$$B + H\text{-}C\text{-}C \longrightarrow BH^+ + C\text{-}C^-.$$

Figure 5

Kinetic isotope effects at 25°C for acid- and base-catalyzed reactions as a function of z, a parameter indicating the extent of proton transfer in the activated complex. ——, from Ref. 20; points for isobutylene from Ref. 21; – – and –·–, from Ref. 18; points for ϕ_2RCH from Ref. 19.



When the parameter z defining the extent of reaction is zero, the activated complex is the unperturbed hydrocarbon; conversely, when $z = 1$, the proton has been completely transferred to the base. Intermediate values of z specify an activated complex in which the proton is bonded partially to the carbon atom and partially to the base. Shown in Figure 5 are values of k_H/k_T as a function of z calculated for the two bases $^-OCH_3$ and Me_3N. (These were obtained from the published values of k_H/k_D by use of the $\ln(k_H/k_T)/\ln(k_H/k_D) = 1.44$, which the authors state to be accurate to within 2%.) The shapes of the two curves are very similar, with a maximum isotope effect predicted for $z \sim 0.5$, i.e., for a symmetrical activated complex. This resembles the analogous predictions in H-atom transfer reactions.

An experimental example is provided by the methoxide ion catalysis of the reaction [19].

$$(C_6H_5)_2CTCH_2O_3SC_6H_5 + {}^-OCH_3 \longrightarrow (C_6H_5)_2C{=}CH_2 + CH_3OT + C_6H_5SO_3^-.$$

The experimental value corresponds to $z = 0.35$ or $z = 0.51$, near the maximum predicted effect.

Similar predictions have been made [20] for acid catalysis by the hydronium ion, with the model

$$H_3O^+ + C{=}C \longrightarrow H_2O + H{-}C{-}C^+.$$

In this model, $z = 0$ corresponds to the hydronium ion. The results, Figure 5, again indicate a maximum isotope effect with $z \sim 0.5$. In the experimental example, the hydration of isobutylene [21] has a lower isotope effect, so the activated complex must not be symmetrical.

Some other examples of base-catalyzed reactions are displayed in Table 4. With the exception of the last entry, the values of k_H/k_T do not lie far outside the range shown in Figure 5. The proposed explanation for this last case is that there is a large contribution to the isotope effect from tunneling [22].

Table 4

Kinetic Isotope Effects in Base-Catalyzed Reactions

Acid	Base	k_H/k_T 25°C	Reference
Malononitrile	H_2O	1.71	40
Acetophenone	OH^-	16.6	41
Acetone	OH^-	11.6	41
Guaiazulene	H_2O	14.1	42
Trimethylazulene	H_2O	26.1	42
1-Bromo-2-phenylpropane	OEt^-	20.7	43
2-Nitropropane	2,4,6-trimethyl-pyridine	78 (32°C)	22

In conclusion, it is apparent that large kinetic isotope effects are often found for tritium-labeled compounds. In tracer experiments utilizing tritium, observed rate constants could easily differ by an order of magnitude from those for the analogous unlabeled compound. If tritium from a source of HTO at constant specific activity is incorporated into a biological system by irreversible chemical reactions, it will be discriminated against, and the tritium level in the biological system will remain lower than that of the source. Conversely, kinetic isotope effects in the back-exchange to remove tritium after incorporation will favor retention of tritium in the biological system.

ACKNOWLEDGMENT

This research was sponsored by the U.S. Atomic Energy Commission.

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DISTRIBUTION OF TRITIUM BETWEEN THE HYDROSPHERE AND INVERTEBRATES

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Although this presentation concerns observed levels of tritium in invertebrates, the data bear on a larger issue: Under what circumstances might tritium develop sufficiently high specific activities that the radionuclide could become a hazard to man? The term "specific activity of tritium" is defined and used subsequently in this paper as the ratio of the number of atoms of tritium to the number of atoms of hydrogen (protium) in a sample. In the papers cited below, the specific activity of tritium is often stated in synonyms such as μCi/cm^3 H_2O, μCi/g of hydrogen, tritium ratios, tritium units, etc., and consequently it has been necessary to translate such data into a form suitable for making comparisons.

It is reasonable to presume that high levels of tritium (atomic weight 3.017) might be incompatible with life since deuterium (atomic weight 2.015) is lethal in mammals when about 30% of the body water is replaced by D_2O. Tritiated water is also lethal, but the $LD_{50/30}$ of tritium in rodents has been reported to be about 1 mCi/g body weight administered parenterally [2,3]. When these data are converted to atoms per gram body weight, it is seen that 2.1×10^{16} atoms of tritium have about the same degree of lethality as 1.1×10^{22} atoms of deuterium. Since the greater lethality of tritium cannot be explained in terms of special physico-chemical properties resulting from the presence of two neutrons in each hydrogen nucleus [4-9], the cause of death is ascribed to the radioactivity of the tritium. This conclusion is strengthened by the fact that tritiated thymidine, which incorporates almost exclusively into the DNA of cell nuclei, is about 1000 times more lethal to living cells on a curie for curie basis than tritiated water [10,11].

Death from radiotoxicity does not, of course, exclude the possibility that aberrant specific activities of tritium may be found in cells or tissues existing in tritium concentrations well below toxic levels. Reasoning by analogy from experiments which demonstrated that the deuterium/hydrogen ratios in biological systems may be greater or less than the normal of about 150 ppm, it is to be expected the tritium/hydrogen ratios in tissues or cells may also be larger or smaller than the specific activities of the media with which they had been thoroughly equilibrated. For example, Carlbom et al. [12] obtained a higher specific activity of deuterium in water by percolating it over barley that was being malted. The sap and wood of the willow tree are reported to have deuterium/hydrogen ratios higher than the normal of 150 to 156 ppm [13]. Beeswax and honey have 17 to 28 atoms percent excess of deuterium while several other natural products have comparable atoms percent deficits of deuterium [14-17].

Other investigators using an aqueous medium containing both tritium and deuterium have found that only 75 to 98% as much tritium as deuterium is incorporated into the products of certain biosynthetic reactions [5-9]. On the other hand, once a tritium atom is incorporated into a methyl, amino or similar group, it tends to be more firmly bound than is a deuterium or hydrogen atom, and it participates in exchanges less readily [18, 19].

It is clear, therefore, that molecules may be generated in biological systems which, taken en masse, represent a greater or lesser tritium/hydrogen ratio, i.e., an enrichment or deficiency of tritium on the biochemical compounds of the cells. The literature on this subject leaves the impression that the specific characteristics of enzyme-catalized reactions combined with the physico-chemical properties of tritium determine whether an enrichment or deficiency will be found. Further, any such effect might be concealed or not detectible in a tissue or organ whose cells are carrying on large numbers of other reactions unaffected by the physico-chemical properties of the tritium.

Experiments in mammals have shown that the specific activity of tritium in the free tissue water will become essentially the same as that in the animal's water supply, provided that the specific activity of the ingested water is constant for some 7 to 10 biological half-lives. The tritium specific activity of the organic components of the tissues, on the other hand, increases, but does not closely approach the specific activity of the ingested water [20-24]. The tritium specific activities of the organic compounds in the various tissues differ widely as to their relative rates of change and final "equilibrium levels" in response to shifts of specific activity of tritium in the free tissue water [20-24]. The differences appear to be associated in large part with the inherent rates of biochemical and cellular turnover of each tissue.

In a carefully devised experiment on rats where the specific tritium activity of the free tissue water was maintained at a fixed elevated level from the fetal stage through the first six months of life, the specific tritium activities of the organic components of the tissues never exceeded 40 percent of the specific activity of the free tissue water or the drinking water [22]. The analogous data for man consist of one direct experiment on the specific tritium activity of fat and dermis biopsied from a man chronically exposed to a fixed elevated level of tritium in his drinking water [25,26] and the indirect evidence from a fatal case of tritium poisoning [27]. Thus it can be concluded with reasonable assurance that mammalian tissue cells are not likely to enrich their organic components with tritium derived from their water supply.

As far as is known, comparable experiments have not been carried out in which animals were fed a diet whose organic molecules were labeled with tritium at elevated constant specific activities. In addition, no data appear to have been published where the specific tritium activities of the ingested water, foodstuffs, free tissue water and organic compounds of the tissues were compared. No investigator appears to have capitalized on the "natural experiment" that would have been practicable when the population was exposed to increases of tritium in the biosphere of two to three orders of magnitude.

If it is postulated that the absorbable subunits of tritiated carbohydrates, fats and proteins are synthesized into organic cellular components with minimal loss of tritium, tritium labeled foods ought to introduce tritium into the organic molecules more efficiently than would be the case of anabolic reactions carried out in a milieu of tritiated water of equal specific activity.

The original question can now be restated as: Do the lower forms of life, and especially those invertebrates and plants which synthesize the basic foodstuffs eaten by higher orders, exhibit evidence of enrichment of tritium in the organic compounds of their tissues? If lower orders do synthesize compounds with high tritium specific activities, those food chains to which they contribute must be investigated in detail to learn if the enrichment is maintained, reversed, or further increased at each trophic level. In general, opinion appears to be against either tritium enrichment or deficiency, but in view of the data on deuterium, these possibilities cannot be dismissed categorically.

Review of Data

Since the literature provides little data on isotopic fractionation of tritium in invertebrates, the inclusion of data on phytoplankton and plants may be permissible. This information has been collated in two forms, 1) narrative with brief annotations, and 2) tabular according to taxonomic groupings in an Appendix.

The specific activity of tritium is conventionally determined for two fractions of tissues or organisms: 1) An aqueous fraction obtained by relatively mild procedures such as lyophilization, desiccator treatment, or passage of dry heated gases over the sample. This fraction is generally called the free water, free tissue water or loosely bound water. 2) A fraction usually reconstituted by oxidation of the hydrogen released during intense combustion of the desiccated sample. This is often erroneously called the "bound water" or "organically bound water" but it is more properly termed bound hydrogen or organically bound hydrogen. At times the latter may include small amounts of water that were not removed by the physical drying procedures. In both cases it is assumed that the tritium moves exactly as does the ordinary hydrogen so that the tritium/hydrogen ratio in the sample represents the original ratio. There is a fair degree of imprecision in the procedures, and the numbers for specific activities should not be interpreted too literally; the original data rarely are accompanied by standard errors or other statistical treatment. Therefore, the tritium specific activity of an experimental sample should be clearly larger or smaller than that of the reference sample before concluding that enrichment or deficiency, respectively, has been observed.

The earliest investigations demonstrated that tritiated water rapidly entered into and exited from the tissue water of the kidney bean plant [28] and of mayfly nymphs, snails, and salmon fry [29]; full equilibrium concentrations were not reached in either experiment but evidence of a "bound-to-tissue" component could be inferred for the bean plant. The specific activity of tritium in the free water of the leaves, stems, and roots of a growing soybean plant did come into equilibrium with that of the medium [30]. The oil in the seed was said to be labeled with tritium but the specific activity of the oil could not be calculated.

Raney and Vaadia [31,32] demonstrated that tritiated water applied to the roots of the sunflower and tobacco plant is transported into the leaves and transpired to the atmosphere; absorption of rain by the leaf surfaces readily diluted the local concentrations of tritium in the tissue fluid of the leaf. Given enough time for equilibrium, the tritium concentrations in the stalk and leaf water approached that in the nutrient fluid but the specific activities of the bound hydrogen were not determined.

Koranda [33] in 1965 appears to have been the first to demonstrate that the specific activity of bound tritium in a biologic sample could be higher than that of its "loose water." He found this occurred in the tissues of hermit and coconut crabs (and mammalian tissues), and in samples of wood and soil. Soils, as compared to rainfall, had bound specific activities of 10^4 to 10^6. These high specific activities probably represent persistence of a previous high equilibrium established during testing. The bound tritium in the coral-based soil appears to have become a reservoir for tritium moving into the plants and animals.

The high levels of tritium existing in the ejecta surrounding the Sedan Crater at the Nevada Test Site also appear to behave as a reservoir. The specific activity of tritium in the water of the ejected soil must have been in the neighborhood of 10^8 times higher than normal when the tissue-bound hydrogen in field-cured plants acquired the same high range [34]. In contrast, no tritium was measurable in the leaves or wood of trees in a tropical rain forest following dissipation of an experimental pulse contamination of the root system [35]. Brown [36] has pointed out that phases of increased concentration of tritium associated with the cellulose of tree rings can be correlated with the levels of environmental tritium that existed during the time the ring was being laid down.

Cohen [37] is finding that the bound and loosely-bound tissue hydrogen of pond weed (Potamogeton), in the Hudson River below the Indian Point pressurized water reactor have specific activities of tritium up to three times higher than the current tritium/hydrogen ratio of the river water. The specific activities of tritium in the bound and loosely-bound water of the root mat and debris dredged from the river bottom tended to be 10 and 1.5 times, respectively, that of the river. It is suggested that these high specific activities reflect the former higher tritium/hydrogen ratios of the river created by atmospheric testing.

The tritium levels in the free tissue water of cattail plants (*Typha angustifolia*) grown from rhizomes in an artificial fresh water pond spiked with tritiated water never attained equilibrium with the tritium concentration in the pond water. The specific activities of the free water from various parts of the plant relative to the water of the pond ranged from 0.58 to 0.7; the specific activities of the bound hydrogen relative to the pond water after nine months of growth ranged from 0.4 to 0.67 [38]. These data are in agreement with unpublished field studies [39] which indicate that the tritium/hydrogen ratios of the bound hydrogen and free water of the following biota reach an equilibrium with the specific activity of the tritium in their environmental water that does not exceed 1.0: leaves, stems, and roots of the pond lily; leaves and roots of bullrushes; pond weed (*Potamogeton*); single cell phytoplanktons, and green and blue-green filamentous algae. It was also found that the total content of tritium per gram of fresh pine needles decreased in parallel with radial distance from the pond which was a source of tritiated water vapor. Comparable studies on grains, vegetables, and fruits are in progress [39].

Phytoplanktons seem to be the lowest form of plant life thus far investigated. The bound water of unicellular *Chlorella pyrenoidosa* cultured in a nutrient salt solution, with tritiated water added to give a concentration of 1 mCi of tritium/ml, reached specific activities of 45 to 49% of the theoretical equilibrium level after six doublings and remained at this level through subsequent doublings [40]. The bound hydrogen of *Chlamydomonas reinhardti*, also a unicellular algae, equilibrated at 67% of the specific activity of the culture medium [41]. The bound hydrogen of two filamentous algae, *Zygnema* [41] and *Pithophora* [38], and of *Lemna* or duckweed [41] equilibrated at 0.72, 0.98 and 0.81, respectively, of the specific tritium activity of the medium.

Two species of snails, *Lymnaea reflexa* and *Helisoma trivolis*, feeding for 180 days on algae growing in tritiated water in laboratory tanks, incorporated higher levels of tritium into their bound tissue water than snails living on normal food plus tritiated water [41]. Also, snails raised from eggs hatching in these tanks developed higher specific tritium activities than did their parents. The observed ranges of specific activities for adults were 0.5 to 0.67, and for the hatchlings 0.6 to 0.9.

The "organically-bound" tritium of *Lymnaea* and *Helisoma*, living in closed tritiated microcosms which were producing tritiated foods, equilibrated at 0.8 to 0.9 of the specific activity of the media. It appears, however, that about 80% of this organically-bound tritium was loosely bound, or at least readily exchangeable, even after eight weeks of equilibration. When the snails were placed in fresh water for three hours, this organically-bound fraction reestablished a specific tritium activity that was approximately 0.2 of the former equilibrium [42].

The water flea (*Daphnea galeata*) grown in laboratory tanks in tritiated water with tritiated algae for food, gave evidence of accumulation of tritium [41]. The specific activity of tritium in the dry tissue relative to the water medium was 1.02 and the specific activity of tritium of the dry tissue relative to the specific activity of the food was 4.3. The fairy shrimp (*Eubranchipus vernalis*) growing in the same medium had respective ratios of 0.2 and 0.6.

The larvae of artificially spawned rainbow trout (*Salmo gairdneri*) when placed in fresh circulating tritiated water rapidly took up tritium for the first one to two days and then maintained an equilibrium for the rest of the 25 ± 10 days of embryogenesis [43]. The bound tritium accounted for approximately 20% of the total bound plus aqueous tritium of the eggs throughout the duration of exposure. When placed in fresh water both the specific activities of the bound and aqueous tritium fractions fell rapidly to 0.1 to 0.3% of their former equilibrium levels, respectively. Both these levels then continued to decrease very slowly for the next 17 days or more [43].

The tissue fluids of the freshwater clam (*Anodonta nuttalliana*) and crayfish (*Astacus sp.*) equilibrated quickly with the tritium levels in the water of an artificial pool [38]. If the specific activity of the bound hydrogen of the tissue fluid of the clam is considered to be in equilibrium with the concentration of tritium in pool water, then the viscera of the clam had the highest specific activity, 0.53, with the gills next at 0.43, and the mantle, muscle and calcareous tissues approximating 0.3. The specific activity increased with length of residence in the tritiated pool; the temperature of the pool was also increasing during this time. The crayfish followed this same pattern.

Blue-fin crabs and oysters in Chesapeake Bay have been sampled at quarterly intervals with the general finding that the tritium ratios of the free water and bound hydrogen from these animals were in equilibrium with, or less than, the specific activities of the surrounding waters [44].

Data on the uptake of tritium by insects are very few. Blaylock and Allen [45] have noted in a progress report that the specific activity of the free water of *Chironomus riparius* is equivalent to that of the pond in which they lived. Crickets reared on tritiated water had the same free water and bound tritium specific activities as the water.

Discussion and Summary

There seems to be more than minor disagreement as to what constitutes "volatile" and "bound" hydrogen, and to some extent the disagreement may be traced to the techniques employed to collect these fractions. There do not seem to be any clear inherent biological demarcations in the degree of fixation or binding. At least three processes are involved: a) absorption, b) exchange and c) firm chemical bonding by synthesis. Considering the variety of colloidal surfaces and complex molecules in a cell, a continuum from volatile to fixed is to be expected. No solution to this problem is offered. Rather a plea is made that those interested in this field come to agreement on a procedure or technique that will provide the most scientifically meaningful cut(s).

The concept of "specific activity" sometimes appears to be employed differently by investigators and its intended meaning within the context of a report is not always clear. With reference to tritium research, the term should be used to express the ratio of the number of atoms of tritium to the number of atoms of hydrogen in a sample. Often this is a clumsy number, difficult to calculate, and for some purposes an unnecessary calculation. However, when the units attached to the numerator and denominator of the ratio become complex and too remote from atoms per g or atoms per ml, a rapporteur has difficulty in arriving at a common denominator for comparing data.

The data collected above, with one exception, indicate that invertebrates do not enrich the organic fractions of their tissues with tritium. Instead tritium appears to be discriminated against. The exception, *Daphnia*, is puzzling and warrants further study. Investigators have noted that tritium is difficult to control technically in the laboratory and it turns up unexpectedly. Investigators working near reactors, or laboratories making tritiated compounds, accelerator targets and the like, should be prepared for the rare, exceptional datum.

Samples of biologic materials originating between about 1954 to 1968 are quite likely to have elevated specific activities of tritium in their bound hydrogen or water of combustion. The environmental levels of tritium during these years were as much as 10^3 times higher than present levels and samples of biological materials synthesized during that period must be interpreted accordingly.

On the whole we seem to be well into the problem stated. However, it would be useful to have additional data on those invertebrate materials which man consumes more or less directly. The specific activities of foods such as nuts, grain seeds, vegetables, shellfish, crabs, seaweed and even hens' eggs would be very illuminating, especially if the specific activities of the media in which these foods grew or were generated were also known.

APPENDIX

Specific Activity of Tritium of Sample Relative to the Specific Activity of the Medium

Phytoplankton		
Unicellular		
Chlorella pyr. [40]	Bound	0.45-0.5
Chlamydomonas	Free	1.0
rein. [41]	Bound	0.67
Filamentous		
Zygnema sp. [41]	Free	1.0
	Bound	0.72
Pithophora sp. [38]	Bound	0.98
Algae in general [39]	Free & Bound	1.0

APPENDIX
(Continued)

Zooplankton		
Water flea (*Daphnia g.*) [41]	Dry tissue	1.02
	Dry tissue/ Tritiated food	4.03
Fairy shrimp (*Eubranchipus v.*) [41]	Dry tissue	0.2
	Dry tissue/ Tritiated food	0.6
Larvae of trout (*Salmo g.*) [43]	Free water=0.8 of total cellular Bound hydrogen = 0.2 of total cellular	~1.0 (?)
Mollusks		
Chesapeake Bay oysters [44]	Free water & Bound hydrogen	~1.0
Fresh water clams (*Anodonta n.*) [38]	Tissue fluid	~1.0
If organically-bound tritium of tissue fluid		= 1.0
the organically-bound tritium of viscera		= 0.53
the organically-bound tritium of gills		= 0.43
the organically-bound tritium of mantle the organically-bound tritium of muscle the organically-bound tritium of calcareous tissue		= 0.3
Snails in general [29]	Free tissue	0.95
Helisoma trif.		
Tritiated microcosm [42]	Loosely-bound	0.8-0.9
Tritiated open tank [41]	Free tissue water	~1.0
	Adult's dry tissue	0.5
	Hatchling's dry tissue	0.7
Lymnaea refl.		
Tritiated microcosm [42]	Loosely-bound	0.8-0.9
Tritiated open tank [41]	Free tissue water	~1.0
	Adult's dry tissue	0.5
	Hatchling's dry tissue	0.6
Water plants		
Duckweed (*Lemna sp.*) [41]	Free	1.0
	Bound	0.81

APPENDIX
(Continued)

Water Plants (Cont.)		
Cattails (*Typha spp.*) [38]		
Leaves, stems, roots, rhizomes	Free	0.58-0.7
	Bound	0.4-0.67
Bull rushes [39]		
Leaves, roots	Free/Bound	<1.0
Pond lily [39]		
Leaves, stems, roots	Free/Bound	<1.0
Pond weed [39]		
Total plant	Free/Bound	<1.0
Pond weed (*Potamogeton*)	Bound	up to 3.0
Surface parts [37]	Loosely bound	up to 1.1
Pond weed (*Potamogeton*) [37]		
Dredged-up root mat	Bound	up to 10.0
Other flora		
Red kidney bean [28]		
Leaves, stems	Free	0.45-0.65
	Bound	1-2% of free
Soybean [30]		
Leaves, stem, roots	Free	0.99-1.04
Tobacco (*Nicotiana*) [32]	Free	0.6-0.95
Helianthus [31,32]	Free	0.9
"Field-cured weeds" at Sedan [34]	Bound	up to 10^6
Wood samples at Pacific Prvg. Gd. [33]	Bound	up to 10^4-10^6
Cellulose of tree rings [36]	Bound	"Proportional"
Grains, fruits, vegetables [39]		
In progress	Free/Bound	<1.0
Pine needles [39]	Bound	<1.0
Insects		
Mayfly nymphs [29]	Free	0.95
Chironomus rip. Midges [45]	Free	~1.0
Crickets [45]	Free/Bound	~1.0

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BIOKINETICS OF ENVIRONMENTAL TRITIUM

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The Environmental Protection Agency (EPA) is involved in evaluation of the environmental aspects of tritium. A review of the efforts of EPA in this field was presented by Moghissi and Carter [1]. The ultimate purpose of all these efforts is to determine possible dangers to people and the environment as a result of the tritium released by nuclear explosives, nuclear reactors, and other nuclear facilities.

Tritium is discharged into the environment in elemental, aqueous, or organic forms. There have been numerous discussions regarding the importance of environmental tritium release in a form other than water. Aside from local problems associated with the release of the tritiated organic compounds, they occur most frequently in either the elemental or aqueous forms of tritium.

Throughout the history of tritium hazard evaluation, emphasis has been on exposures to tritiated water. Associated radiation doses have been calculated with the assumption that ingested tritium remains in the body water until excreted. Several studies with animals and people have indicated that significant amounts of tritium are incorporated from body water into organic constituents of the body and are excreted with half-times much longer than that for body water [2-5]. Moreover, tritium taken up by plants as tritiated water is also incorporated in plant tissue, where the turnover rate is different from that of water in the plants [6,7].

Tritium released into the environment would, to some degree, be dispersed throughout all the hydrogen of that environment. The biological half-time of tritium in the species of primary interest, man, would depend on all the complexities of metabolism in various parts of the environment and man. It should be emphasized that consideration of chemical form and environmental interactions is not a new concept in radioisotope hazard evaluation; however, in the specific case of tritium these factors have largely been ignored.

We are primarily interested in the evaluation of the radiation dose to man which results from environmental release of tritium. This evaluation can be carried out by using a dynamic approach, a steady state assumption, or a combination of the two.

Dynamic Approach to Dose Estimation to Man

This approach is particularly useful for evaluation of the hazard to workers of a nuclear facility or to the population in the immediate vicinity of a source of tritium release. Except for cases where tritiated organic compounds may be released in large quantities, it can be safely assumed that the chemical form of the released tritium is water. Any elemental tritium would either escape the area rapidly or be oxidized to water. Although tritium enters the ecological cycle in the form of water, it soon becomes a part of all hydrogen-containing compounds. An adequate evaluation of the tritium movement through various chains of the ecological cycles would thus require the knowledge of kinetics of reactions between chemical compounds of the living organisms. This would include both reaction orders and reaction rates. A solution to these requirements in the foreseeable future is not anticipated. Consideration of the movement of water only, ignoring the entrance of tritium into the food chain and into human tissue, is an unsatisfactory approach because it underestimates the radiation dose by an unknown amount.

Figure 1

Simplified Model for Transport of Tritium in the Environment

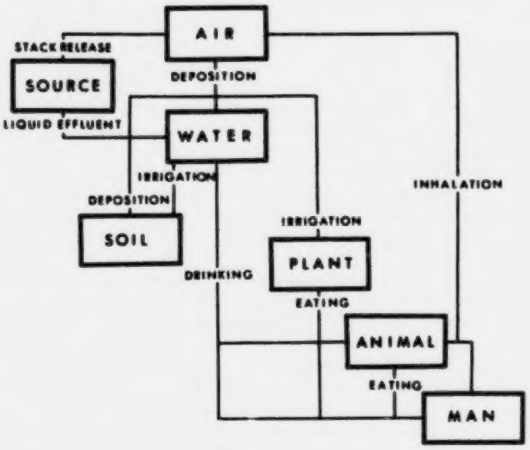
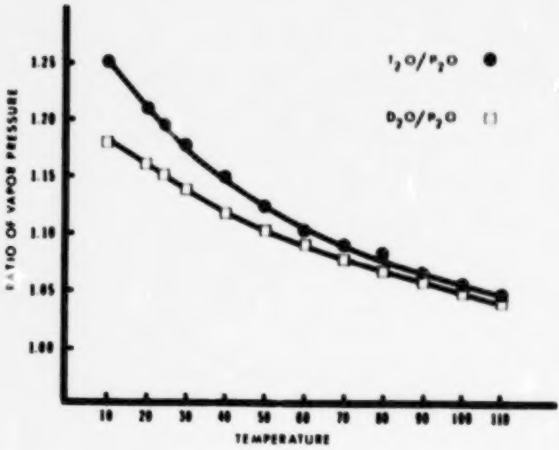


Figure 2

Ratio of Vapor Pressure of P_2O/T_2O and P_2O/D_2O after Jones



It may be advantageous to briefly discuss, at least qualitatively, the dynamic approach. To facilitate the discussion, Figure 1 is used. Tritium released from the stack or as liquid effluents is deposited on the soil by various mechanisms such as precipitation and irrigation.

The behavior of tritium in soil is discussed extensively in this book and thus need not be repeated here. Based on the work reported by Munnich [8] and on results obtained in our Laboratory, tritium deposited on soil migrates by a mechanism comparable to adsorption chromatography. That is, each time the band migrates, its width is increased, but complete mixing is surprisingly not as rapid as one would expect. This is, however, analogous to the behavior of tritium in the oceans, as discussed elsewhere in this book. The movement of tritium in the air and water is beyond the scope of this paper. The behavior of tritium in plants is, however, of particular interest and deserves consideration.

There is sufficient evidence that tritiated water introduced into the aqueous component of a plant is rapidly mixed with the entire water so that the specific activity of water taken from any part of the plant is essentially the same. This is comparable with equilibrium conditions in mammals which have been reported repeatedly and which are discussed in this book. The dynamic behavior of tritium in organic compounds of a plant is complicated and depends upon a number of parameters, such as the nature of species, temperature, growth rate, etc.

These considerations are also valid to some extent for animals, and particularly for human beings. If tritium is introduced into the human body, a fraction of it is excreted with a half-life of approximately 10 days. In addition, there are at least three additional half-life components in man [9]. These are less than one day [9], approximately 30 days [4,9,10,15] and a year [9,10,15] or more. An equation describing the four-fraction excretion would be:

$$A = s\,e^{-t\ln2/Ts} + w\,e^{-t\ln2/Tw} + i\,e^{-t\ln2/Ti} + l\,e^{-t\ln2/Tl}$$

where Ts, Ti and Tl refer to the half-lives of tritium denoted as short, intermediate, and long, respectively, and Tw refers to the turnover rate of body water. The values for s, w, i and l represent fractional activity coefficients of each of the corresponding half-lives and vary with the compound introducted into the human body.

It is extremely difficult to estimate the dose under a dynamic condition with any degree of precision due to the lack of knowledge of fractional activity coefficients for animal and plant life. In the environment, these coefficients would be affected by various ecological factors in the animal food chains. The above equation is for excretion of tritium after all exposure to tritium ceases. Other equations could be written for the uptake into the various components or compartments during tritium intake. A set of uptake and elimination equations has been written and the coefficients experimentally defined for rats exposed to tritiated water [3]. However, such equations provide only a lower limit for dose calculations when the entire environment, including the food supply, is contaminated with tritium.

In experiments with animals [11,12] exposed to tritiated water only, it has been found that about 30% of the protium associated with organic compounds exchanges rapidly with tritium in body water. In addition, about 25% of protium in the organic form exchanges with tritium at much slower rates, perhaps through metabolic reactions [3,12-14]. The remainder of body protium appears relatively inaccessible to tritium exchange from body water, possibly reflecting body utilization of intact food molecules. The specific activity of tritium in plants should reflect that in the water and, therefore, the plant food of animals also would be tritium labeled. Thus, the specific activity of organic body protium in animals should approach that in environmental water provided no concentration mechanism for tritium occurs.

Steady-State Equilibrium Approach to Dose Estimation to Man

If the radiation dose to the public is sufficiently small and an averaging process can be applied, the problem is substantially simplified. Strictly speaking, a steady-state equilibrium would require a constant, specific activity of tritium in terms of pCi/g H or an equivalent unit in water. Presumably, plants and animals would establish a steady-state equilibrium with the hydrosphere, whereby the specific activities of tritium in various components of the biological system would be constant. It is unfortunate that the importance of such a simple approach has been recognized only recently.

Because tritium enters all components of biological systems in substantial amounts, the time has come to define a reasonable estimate of tritium concentration in the environment or components thereof under steady-state conditions of contamination. Therefore the following rule is proposed:

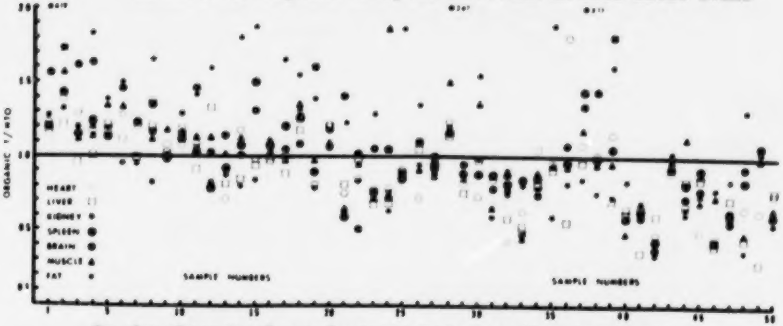
> *Under steady-state conditions, the ratio between different hydrogen isotopes in various components of the biological system is essentially constant.*

It would be of interest to evaluate the validity of this proposed rule using presently available data.

There is a possibility that an isotope effect occurs in the passage of water through the membranes of a plant. It is probable that transpiration is associated with an isotope effect. Figure 3 shows a graph of the data reported by Jones [16] indicating the ratio of vapor pressures of ${}^1H_2O/{}^3H_2O$ and ${}^1H_2O/{}^2H_2O$. Under environmental conditions one deals with ${}^1HTO$ rather than T_2O, so that in the absence of any other data, the D_2O data may be applied. It is important to point out that the isotope effect is temperature dependent; thus one has a better chance to observe it in plants in cold climates.

Figure 3

The Ratio of Organic to Aqueous Tritium in Deer after Evans



In the absence of conclusive tritium studies in plants, deuterium studies may be used to evaluate possible isotope effects. Wershar et al. [17] have shown that, under environmental conditions, water samples extracted from various parts of the plants may exhibit different deuterium concentrations. While deuterium concentrations in the water samples extracted from phloem, xylem and root were only slightly different, these values deviated in some cases by 5 to 6% from that measured in the aqueous portion of the leaves. This may be explained by differences in the vapor pressure of H_2O and HDO; similar differences were also observed in the water collected from various depths of soil. The surface of the soil was somewhat concentrated in deuterium indicating the cause of enrichment to be the evaporation of water which occurs primarily on the soil surface. Assuming an equilibrium between the specific activity of tritium in water and that in organic molecules in the plants, a fractionation between the tritium activities in leaves and in other parts of the plants would occur which could amount to 30% differences for organic tritium, if the deuterium values of Wershaw are extrapolated to tritium. Controlled experiments under steady-state conditions are required to confirm the validity of these observations.

The data reported by Chorney et al. [7] indicate an agreement between the aqueous and organic parts of the plant. Results from certain plant samples collected by our Laboratory from the Rulison area, and preliminary results from controlled experiments in this Laboratory, show a reasonable agreement with this postulate.

A study dealing with tritium in organic and aqueous components of mammals (deer) has been reported by Evans [18]. As shown in Figure 3, large variations were observed among tritium concentrations in different tissues. The data of Evans [18] and Martin and Koranda [19] do not contradict this proposed rule. Although large discrepancies between individual tissues were observed, these can be explained by the fact that the steady-state condition did not occur during exposure. For example, the deer studied by Evans were free to migrate throughout a large area with different tritium specific activities. Although this freedom of migration was not available to the kangaroo rats studied by Martin and Koranda, the change in the environmental tritium levels with time could account for the differences observed.

It should be of interest to note that improvements in tritium methodology have made it unnecessary to study this problem using animals raised under elevated tritium conditions. For this reason, studies with human tissues are in progress in our Laboratory and the results will be reported in due time.

The possibility of an isotope effect mentioned for plants has been observed in humans [20]. Tritium analysis of condensed water from breath indicates approximately 10% lower specific activity than the analysis of tritium in body water, which is in good agreement with the data of Jones shown in Figure 2. Many investigators report the occurrence of a relatively small carbon isotope effect [21]. It is expected that this effect is magnified with hydrogen. In addition there is no question that between protium and tritium an isotope effect of varying magnitude occurs in every reaction which is not completed, and almost all biological reactions fall into this category. This rule postulates that although a large isotope effect may occur in each one of the reactions involved in the uptake, utilization, and excretion of tritium in either aqueous or organic forms, the net result is negligible.

The only study known which by implication violates this proposed rule has been reported by Cloud et al. [22], and Krichevsky et al. [23]. These authors have shown that specific bacteria, capable of converting formate to hydrogen, fractionate hydrogen isotopes with separation factors comparable to those obtained during the electrolytic process. Presumably, hydrogenous materials remaining in the aqueous phase will be enriched with heavier hydrogen isotopes after prolonged bacterial activity.

No other instance is known in which the tritium specific activity in organic portions of plants or animals exposed to HTO exceeds that in the body water of the organism during some period of its life. Indeed, for animals the data indicate the food and water supplies must be tritiated in order for body water and tissue tritium to reach specific activity equilibrium.

The steady-state equilibrium approach is not as conservative as it first appears. Specific activity of hydrogen in people is assumed to be the same as in other components of their environment. The data discussed previously clearly show that the specific activity of hydrogen in plant or animal tissue can substantially exceed that in tissue water. The relationship between tissue water and organic tritium depends on the tritium exposure history, as for most other radionuclides which have been studied in the past. With active tritium incorporation into and elimination from tissues of animals, the reasonable average specific activity in organic components would be that in the water in the general environment. If levels in the environment fluctuate widely or approach levels where the associated radiation dose becomes of concern, then easily attained and analyzed environmental water samples are not sufficient. Detailed analyses of organic tritium in foods as well as body water are required to evaluate human radiation doses. If the environment should become contaminated to this extent, then the fractional activity coefficients as well as detailed information on ecological cycling of tritium for dynamic calculations are needed. If released tritium is not thoroughly dispersed in the environment, then the environment must be studied in terms of equilibrated hot spots, dynamic studies must be performed, or the two approaches may complement one another. To the extent that equilibrium is not established throughout the environment, specific activities will be higher in some locations. If these locations lie on human food pathways, then potential hazards may be greater than for complete equilibrium.

Conclusion

The proposed rule is that under steady-state conditions the ratio between different hydrogen isotopes in various components of biological systems is essentially constant. Further studies are needed to test the validity of this proposed rule. Until confirmed, a basic prerequisite to general use of this proposed rule is that the radiation dose to the public is sufficiently small so that an averaging process can be used for environmental tritium. The underlying concept for this rule is not new as the EPA and its predecessor organizations have used this assumption for dose estimates related to environmental tritium. The appropriate equation is as follows [23]:

$$H = 0.1\ QC$$

where H is the dose equivalent in mrem/a, Q the quality factor, and C the tritium concentration in body water in pCi/ml.

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TRITIUM TURNOVER RATES IN MAMMALS

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The title of this paper is necessarily an oversimplification of the intended topic, for the reason that tritium as such does not have a meaningful turnover rate. Instead, we shall be concerned with the turnover rates for various tritium labeled compounds, and these will be considered in relation to environmental sources of exposure and in relation to the problem of the radiation dose from internally distributed radioactivity.

Much of the material pertinent to this subject has been reviewed recently [1-4]. The older literature has been very thoroughly and competently reviewed by Feinendegen [5]. Another more recent review by Woodward [6] is a good source of references and gives a critical review of many key topics.

Although any compound that contains hydrogen may be labeled with tritium, by far the most important tritium containing compound in the environment is water. In part by virtue of its abundance in the body, water is also the most important form of internally-distributed tritium and is the major catabolic product of most of the labeled molecules which enter the body.

Therefore, we shall begin with a discussion of the turnover of water. Tritium labeled water may enter the body through several routes: by respiration, by ingestion, or by diffusion through the skin. The early work of Pinson [7], Pinson and Langham [8] and Delong *et al.* [9], and more recently by Osborne [10] indicate that diffusion through the skin is a surprisingly efficient route of tritium intake, the rate of uptake from tritiated water in the air being about equal for respiration and for diffusion through the skin. In either case, the tritium uptake per minute is equal to the tritium activity in 10 l of air.

The metabolism of water in representative animal species has been reviewed by Feinendegen [5]. When the body is treated as a one compartment system, for the mouse the turnover half-time is 1.13 ± 0.14 days. At the other extreme is the kangaroo rat, maintained on dry pearled barley only, with a half-time of 11.8 ± 3.0. Other species have intermediate figures, with that for normal man being taken as eight or nine days, but with variations cited [5] ranging from less than seven to more than 10 days. Other reports [10-12] include values ranging from 4 to 20 days.

For human beings, it requires quite a stretch of the imagination to consider anything so complex as the human body as a single compartment. Edelman [13] has reviewed the physiology of body water. There are some six main subdivisions of the body water, with about 55 percent being intracellular and 45 percent extracellular, the latter being subdivided into plasma, interstitial lymph, connective tissue and cartilage, bone, and transcellular fluid. The transcellular category includes water in the exocrine glands, biliary tree, kidneys, eyes, cerebrospinal fluid and the gastrointestinal tract. No appreciable region of nonexchangeable water is known to exist. Despite the complexity of the system, however, studies show that the tritium disappearance curve may be reasonably closely represented by only two or three exponential components, one with a half-time of 10 days and the other two being about 30 to 300 days. Such representation is only descriptive, and the individual components cannot be assigned to individual compartments of the system. With this reservation, the slower components evidently represent exchangeable hydrogen in organic compounds. Tritium does not have to enter the body in some exotic form to get into the slower pools. Some of the tritium that enters the body as water exchanges with the hydrogen in the long-lived pools or spaces in the body. The ICRP Committee II report [14] used a half-time of 12 days [15]. The various models and curve-fitting equations that have been used are compared in a later report [16]. Siri and Evers [17] found 7 percent of the injected label in dry tissue solids one day after the injection of a large single dose. Smith and Taylor [18] have examined the biochemical pathways by which tritium gets into organic compounds in the body, particularly adipose tissue, fatty acids, nucleic acids and carbohydrates. Most acute experiments indicate that only a few percent of administered tritium finds its way into the longer-lived components.

Several studies involving chronic exposures are pertinent, however, such as Evans' [19] study of 52 deer exposed to tritium from a tritium-producing installation. The tritium concentrations in the body water of these deer ranged from 4.2 to 54.5 pCi/ml. Seven tissues, heart, liver, kidney, spleen, brain, muscle, and fat, were freeze dried and assayed for organically bound tritium. It was estimated that about 0.85 of equilibrium specific activity had been attained. For radiation dose calculation, it was assumed that eventually all of the whole body hydrogen could become uniformly labeled. Pursuing this line, with the hydrogen content of standard man being 7.0 kg, the relationship that 1 μCi/l in body water could give a total body burden of 63 μCi was obtained, in contrast to the 43 μCi obtained if only water is considered.

Koranda et al. as cited in Robison and Anspaugh [20], obtained somewhat similar results with kangaroo rats that lived for several generations in the elevated tritium environment surrounding the Sedan Crater at the Nevada Test Site. Here, the activity per gram of hydrogen in lyophilized residues of six organs averaged 1.5 times as high as that in body water. On the basis of these results, they predict a dose in man 1.8 times that obtained using the the ICRP [14] model. However, in evaluating the hazards from Project Rulison, Robison and Anspaugh [20] point out that these data contrast with the data of Thompson and Ballou [21] who exposed mature female rats to constant levels of tritiated water, mated them after six weeks of exposure and continued by maintaining the offspring on the same exposure level for six months. Here the tissue components showed 20 to 30 percent of the activity in the body water during the exposure. This was interpreted to mean that only some 20 to 30 percent of the organic hydrogen in the body comes from water, and that the higher results mentioned above for deer and kangaroo rats indicate that these animals obtained some of their ingested tritium from food as well as from water. The Rulison project is also discussed by Wruble and Andrews [22]. A study under way by Moghissi [23] involving rabbits being fed on alfalfa grown with tritiated water may be expected to further clarify the situation.

Jaworowski and Bilkiewicz [24] have demonstrated the incorporation of tritium in hair, and suggest that hair may be used for the evaluation of internal contamination of the body with tritium.

A question frequently arises concerning the possibility of an isotope effect resulting in a compartment in the body having a specific activity of tritium higher than that in the food or water. This is a legitimate question because the mass of the tritium atoms is so much greater than that of the protium atom, and it is known that the reaction rate of many tritium labeled compounds is detectably slower than that of their unlabeled counterparts. Weston [1,25] documents the mechanisms by which the kinetic isotope effect can involve molecules of biological interest. To be of radiobiological significance, however, such an effect would have to influence a large pool of hydrogen or result in a very high increase of specific activity in a key molecule.

The direction that the isotope effect takes is usually to make a reaction involving tritium slower than with protium. This can result in the piling up of tritium in the affected reactant, but by the same process, the reaction products tend to be depleted in tritium. The process continues until at equilibrium in a chain reaction such as $\rightarrow A \rightarrow B \rightarrow$, with isotopic discrimination retarding the flow of tritium from A to B, the specific activity increase in A is exactly balanced by the discrimination factor, so that the specific activity in the inflow into B is the same as that into A. As an illustrative model if X_0, X_A, and X_B are the specific activities in the inflow, compartment A and compartment B, respectively, and k_A and k_B are the turnover rate constants for compartments A and B, and f is the isotope discrimination factor for A → B, equations 1 and 2 describe the steady state situation. It should be noted that in steady-state systems, the tracer behaves as if the specific activity were the driving force, giving an appearance of first order kinetics, regardless of the actual reaction order [26]. For nonsteady state situations, the analysis is usually much more complicated than this simple example.

$$\frac{d\,X_A}{dt} = k_A X_0 - k_A\,f\,X_A \tag{1}$$

$$\frac{d\,X_B}{dt} = k_B\,f\,X_A - k_B\,X_B \tag{2}$$

The solutions for the system of equations (1) and (2), assuming that at $t = 0$, $X_A = X_B = 0$, but X_0 is finite are:

$$X_A = \frac{X_0}{f}\,[1 - e^{-k_A f t}] \tag{3}$$

$$X_B = X_0 \left[1 - e^{-k_B t}\right] + \frac{k_B X_0}{k_B - k_{Af}} \left[e^{-k_B t} - e^{-k_A f t}\right] \qquad (4)$$

At $t = \infty$

$$X_A = X_0/f \qquad (5)$$

and

$$X_B = X_0 \qquad (6)$$

Thus, there may be intermediate compounds with increased specific activity without this increase being propagated downstream in the reacting system, because the discrimination factor and the specific activity increase are balanced at equilibrium. A tissue containing a compound having an increased specific activity may thus receive a higher radiation dose than would be calculated from input and output specific activities.

In the case of water, a significant site at which isotopic discrimination occurs is the lung, where there is a change from the liquid to the gas phase. In experiments with animals, Mazrimas and Hatch [27] found the specific activity of tritium in expired water vapor relative to tritium in tissue water to be 0.44 in kangaroo rats and 0.64 in mice. The effect of isotopic discrimination in the lung on the specific activity of body water depends on the fraction of water excretion that is handled by the lungs. If X_0 is the intake specific activity, X_1 the body water specific activity, k the rate constant for the total turnover of water, k_L the rate constant for water excretion in the lung, and f the isotopic discrimination factor, then at equilibrium:

$$X_1 = \frac{k}{(k - k_L) + k_L f} \; X_0$$

For example, if 25 percent of the water excretion is via the lung, and f = 0.5,

$$X_1 = \frac{X_0}{0.75 + 0.125} = 1.14 \; X_0$$

Thus, isotopic discrimination in the lungs could lead to an increased specific activity in the body of the order of 15 percent above that in the ingested water. Experimental data are needed to determine whether the animal results may be extrapolated to man. As explained above, some tissues may have even higher specific activities of tritum.

A possibility often discussed, but never shown experimentally except for transient situations, is that, through the isotope effect, tritium could be selectively concentrated in some compound at specific activities higher than those in water. However, some tritium labeled compounds are well known to concentrate in locations that enhance their radiobiological effectiveness. In particular, tritiated thymidine finds its way into the DNA molecules, which is, of course, about as radiation sensitive a site as can be found. Wade and Shaw [28] have recently studied this problem in mice. After 24 hours, the average retention of tritiated thymidine was 51 percent, of which eight percent was in the nonvolatile fraction. It was concluded that tritiated thymidine gets into DNA by a process not related to preparation for mitosis. The activity was concentrated in about five percent of the cells in the tissues studied. Extrapolating to man, it was estimated that ingestion of 1 mCi of tritiated thymidine would give a dose of 1.25 rem to the cells in which it concentrates, compared to 20 mCi of tritiated water required to deliver the same dose. This result is important in the consideration of laboratory exposures to tritiated thymidine, but not in the more general tritium exposure situation.

In conclusion, the available data on tritium turnover rates indicate that for acute exposure, calculation of the radiation dose using the ICRP [15] half-time of 10 days is reasonable, but for chronic exposures the total dose should be taken as about 1.5 times that calculated for water because of the incorporation of tritium in organic compounds.

ACKNOWLEDGMENT

The assistance of M. Comstock in preparing the bibliography is gratefully acknowledged. This research was supported by the U. S. Atomic Energy Commission.

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TRITIUM EXCRETION IN MAN UNDER TROPICAL CONDITIONS

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Personnel working in and around tritium contaminated areas inevitably take small quantities of tritium into their body by skin absorption, inhalation, and ingestion as oxide, a form in which it is frequently encountered. Since tritium is distributed uniformly in all body fluids, it gives uniform exposure to the whole body and doses from the internal source must be added to those from external sources to compute total whole body exposure. The internal dose from tritium received by an individual can be calculated if the biological half-life is known. A number of workers have found the biological half-life for tritium for a standard western adult and it is on these findings that the maximum permissible concentration levels are based. Since biological half-life of tritium depends on ambient temperature and food habits such as water intake, it is considered doubtful if the values reported in the literature for a standard western man may be applied to persons living in tropical countries where conditions could be quite different. Earlier studies by Venkataraman et al. [1], ICMR [2] and Raghunath and Soman [3] have shown that the daily water intake through fluid and food alone for an Indian adult is 4.1 liters and that the seasonal variation ranges between 3.5 and 5.5 liters a day. Based on 4.1 liters water intake, Feige [4] has pointed out that for an Indian adult, the biological half-life for tritium should be less than 6.5 days, as compared to 10 days reported by ICRP. In view of this and the increasing interest in tritium hazards, it was thought advisable to establish the biological half-life for tritium under local conditions.

Table 1

Summary of Data and Results

Case No.	Age Year	Body weight kg	Average daily water intake l	Average daily urine passed l	Time required for tritiated water to come into equilibrium with body water h	Estimated body water: from dilution of tritiated water taken l	Estimated body water: as % of total body weight	Biological half-life: from urine samples days	Biological half-life: from exhalation samples days
1	28	76	2.9	2.0	2-3	43	58	6.2	6.9
2	35	53	2.1	1.1	1-2	30	57	8.9	9.3
3	28	60	2.9	2.7	1-2	34	57	4.9	4.4
4	35	57	1.6	1.2	1-2	38	67	7.3	8.9
5	31	47	1.4	0.6	1.5	35	74	5.8	6.4
6.	36	57	-	-	2-3	36	63	7.2	-
7	39	60	2.2	0.8	2-3	35	58	6.3	-
8	34	46	-	-	1-2	29	63	6.2	-
9	32	83	-	-	2-3	48	58	7.1	-
10	32	45	0.93	1.1	2-3	26	58	5.9	-
11(a)	34	62	1.7	1.6	2	33	53	5.9	-
11(b)	29	60	1.3	1.3	2	27	45	8.5	8.5
						Average	59	6.7	7.4

Method and Materials

Eleven staff members volunteered for the acute exposure to tritium. It was planned that no subject would receive an amount exceeding one maximum permissible body burden prescribed by ICRP. In order to study the water intake habits of the volunteers, they were requested to keep a record of volume of water, tea, milk and other soft drinks consumed daily during the period of study and of the volume of urine they passed. The data collected is summarized in Table 1.

The experiment was divided into two parts. First, each of six volunteers was given 314 μCi of tritium oxide in 250 ml of drinking water, and urine and exhalation samples were collected. Measurements on these samples confirmed that the specific activity of urine and exhalation moisture is the same. Therefore, in the second part of the experiment, exhalation samples were not collected, and each of six volunteers was given 460 μCi of tritium oxide in drinking water.

The exhalation samples were collected using a coldfinger. The urine samples were distilled immediately after collection and the distillate so obtained was used for measuring tritium content in urine samples.

Both exhalation and urine samples were collected from each subject just before tritium intake and at regular intervals after intake. In order to study tritium assimilation in body water and to determine the time taken for tritium to come into equilibrium with body water, hourly samples were taken for the first day (eight hours). Thereafter, sampling frequency was progressively decreased from twice a day to once a week. Sampling was continued until the specific activity decreased to detection limit. The sampling periods ranged from one to two months.

The samples were counted by liquid scintillation. One ml of sample was added to 15 ml of scintillator solution composed of 100 g of napthalene, 0.3 g of 1,4-bis-2-(5-phenyloxazolye)-benzene (POPOP) and 7 g of 2,5-diphenyloxazole (PPO) dissolved in one liter of dioxane. All the chemicals used were of scintillation grade.

Samples were counted in low background glass vials at 10°C. Efficiency of the system used was 11%, and the lower detection limit with 95% confidence was 200 pCi/ml. The method adopted for determination of internal dose received by an individual was the same as that followed by Lawrence [5].

Figure 1

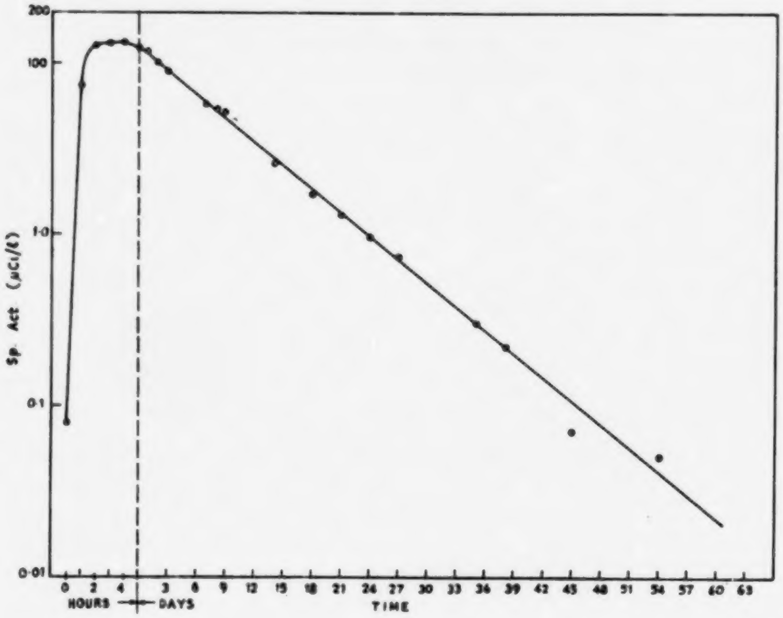
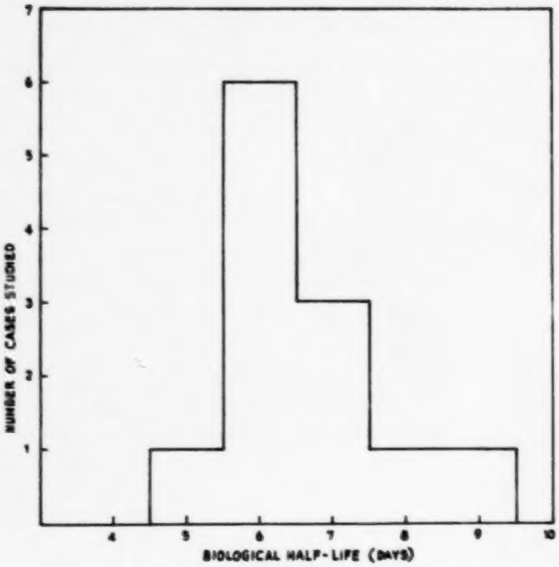


Figure 2

Spread of Biological Half-Lives



Results and Discussion

From the hourly samples collected for the initial period, it is observed that the time for the intake water to come into equilibrium with body water varies between one and three hours, Table 1. A typical case is shown in Figure 1. In the case of one volunteer whose samples were collected at intervals of one-half hour, the equilibrium time was observed to be 1.5 hours. The equilibrium time of one to three hours obtained by us compares favorably with the reported time of about 2.5 hours [6].

Figure 1 gives a representative specific activity vs. time curve for a typical case, and Figure 2 gives the distribution of half-life in the cases studied. The results are summarized in Table 1. Our studies show a spread over four to nine days, with an average value of 6.7 days for the biological half-life of tritium. This spread of four to nine days is small as compared to that of 4.3 to 13 days obtained by Pinson and Langham [7] and four to eighteen days obtained by Butler and LeRoy [8].

The average value of 6.7 days is significantly less than 10 days used in ICRP data and 11.5 days, 9.5 days and 8.5 days reported by Pinson and Langham [7], Richmond _et al_. [9] and Wylie _et al_. [10], respectively, from their studies in the USA. The value of 6.7 days obtained compares well with 7.5 days reported by Foy and Schnieden [11], from their work in Southern Nigeria.

In the present work there was no indication of more than one component half-life for tritium as reported by Lawrence [5], though in both cases, studies were confined to a period of two months.

The biological half-life for tritium varies from individual to individual, and for the same individual, during different seasons. In one case, the biological half-life during winter and summer was 8.5 and 5.9 days, respectively, when the average ambient temperatures were 25° and 33°C. This indicates that, during different seasons, when ambient temperature variations are more pronounced, biological half-life will vary considerably.

Figure 3

Water Intake and Biological Half-Life as Function of Age

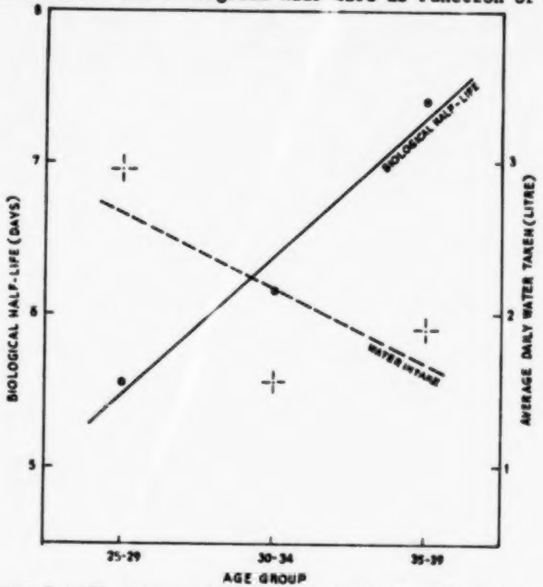


Figure 3 indicates variation of half-life and daily water intake with age. It may, however, be emphasized that these observations are restricted to eleven cases only. Our data indicate that the biological half-life varies directly with the age of the subject, whereas the observations of Butler and LeRoy [8] are just the reverse. The average daily water intake decreases with age, Figure 3, which probably accounts for the observed half-life variation with age. However, it may be pointed out that Raghunath [12], in his physiological studies for Indians, observes that the daily average water intake is almost constant for the age groups between 20 and 40 years.

From the gross activity taken in and the specific activity of urine samples after tritium oxide has come into equilibrium with body water, it is possible to calculate the body water volume for an individual. Table 1 gives the body water for subjects studied; the average is 59% of total body weight, which is in agreement with 60%, 62.2% and 61.8% reported by Wylie et al. [10], Pinson and Langham [7], and Foy and Schnieden [11], respectively. However, values for body water arrived at using Wylie's method [10] were considerably lower (43%). The large discrepancy could be due to the assumption that the total daily water output is 5/3 of daily urine output, which may not be applicable under tropical conditions.

Table 2

Daily Water Output

Case No.	Estimated body water V_t (1)	Biological half-life T_b (days)	Daily water output V(1) $\frac{0.693\ V_t}{T_b}$	Daily urine passed U(1)	V/U
1	43	6.2	4.8	1.95	2.47
2	30	8.9	2.34	1.12	2.09
3	34	4.9	4.81	2.7	1.78
4	38	7.3	3.61	1.19	3.03
10	26	5.9	3.05	1.12	2.72
11(a)	33	5.9	3.88	1.64	2.37
11(b)	27	8.5	2.2	1.3	1.69
				Average	2.3

Using Wylie's method [10] and the biological half-life of different cases studied here, the total daily water output works out to 3.66 liters, cf. Table 2. This deviates substantially from 2.5 liters for the ICRP standard man. Using daily total water output so calculated, the ratio of daily water output to daily urine output was calculated for those cases for which urine output was known. The average ratio works out to 2.3 as compared to 1.67 for a standard man [13]. Out of nine cases, two showed abnormal urine output and were not considered in the above calculations.

Our studies indicate that in this region the average biological half-life for tritium is approximately seven days as compared to 10 days used by ICRP in deriving MPC values. Hence, it is suggested that these values should be suitably revised for application in India.

ACKNOWLEDGMENTS

The authors are grateful to Dr. A. K. Ganguly, Head, Health Physics Division for his keen interest in the present work and to staff members of BARC who participated in this work.

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TRITIATED WATER TURNOVER RATE IN DESERT MAMMALS

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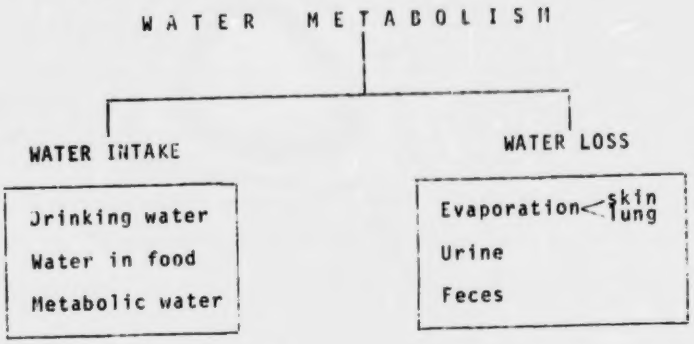
Aridity and intense heat constitute a great challenge to survival of mammals in the desert. It is well known that water metabolism is a function of water intake and water loss. Different avenues of water metabolism are summarized in Figure 1. Water loss is influenced by an animal's heat production plus heat gained from the environment. The accumulated heat must be dissipated to prevent an intolerable rise in body temperature. In a given organism the turnover of energy is related to, or dependent upon, its water expenditure. Examining the physiological responses of animals to the desert, Dill [1] divided desert mammals into two groups: those who use water for thermoregulation, i.e., large mammals, and those who do not use water for thermoregulation, i.e., small mammals.

Figure 1

Different Avenues of Water Intake and Loss



Although it is impossible to separate discussion of water metabolism and of thermoregulation, this paper deals primarily with water turnover rate in large and small desert mammals. Evaluation and interpretation of the data will be made in the light of the facets of temperature regulation.

Determination of daily water exchange between an animal and its environment is tedious and technically difficult to accomplish. The discovery of the usefulness of tritiated water in measurement of total body water by Pace, et al. [2] has offered a practical method for the study of daily water exchange. This technique has been verified in man [3] and in several other mammalian species [4]. Briefly, the technique consists of an injection of a dose of tritiated water. This quickly equilibrates with body water, and the tritium concentration in body water decreases exponentially with time. The turnover rate of tritiated water is calculated from the changes in the concentration of tritium in several consecutive plasma samples. The concentration of tritium in plasma is calculated from the equation:

$$K = (\ln A_o - \ln A_t)/\Delta t$$

where A_o is the activity of tritium at zero time, A_t is the tritium activity at time t, Δt is time and k is the fraction of body water pool turned over per unit of time. The biological half time, T_b is equal to 0.693 divided by k.

The time required to remove a volume of water equivalent to the size of the body pool is termed turnover time, and it is calculated from the equation:

$$\text{Turnover time} = 1/k$$

The total body water is then calculated from the equation:

$$\text{Total body water ml} = \left[\frac{(\text{Std activity, ml}) \times (\text{std dil. factor}) \times (\text{dose volume, ml})}{\text{Activity in ml at zero time}}\right]$$

Knowing k/day and total body water, ml, one can calculate the total water loss per day as:

$$\text{Water loss, ml/day} = (\text{k/day})\ (\text{total body water, ml})$$

Since, under normal physiological conditions, the animal is in water balance (water intake = water loss), the calculation of water loss/day is an index of daily water intake. Studies on T_b of small and large desert mammals as compared to nondesert ones are summarized in Table 1.

Table 1

Biological Half Time, T_b, of Large and Small Desert and Nondesert Mammals

Large Mammals	T_b,day	Reference	Small Mammals	T_b,h	Reference
Man	9.5	4	Round-tailed ground squirrel	92	18
Burro (donkey,ass)			Antelope ground squirrel	72	18
Spring	5.5	This paper			
Summer	3.8		Palmers chipmunk	36	18
Bighorn sheep			Chissel-toothed kangaroo rat	171	18
Spring	6.5	This paper			
Summer	4.5				
Camel			Desert kangaroo rat	284	4
nonlactating	7.5	17	Laboratory white rat	85	4
lactating	5.5	17	Laboratory white mouse	27	4
Merino sheep					
Arid tropics	4.5	17			
Subtropics	3.5	17			
Zebu cattle	3.5	17			

For small mammals among the desert forms, there is a seven- to eightfold range in water turnover rate among those studied, with the two species of kangaroo rats having the least turnover rate and the chipmunks having the greatest (Table 2). This great variability among the desert rodents reflects a wide range of metabolic and behavioral adaptations. For example, some of these species are nocturnal while others are diurnal. They also burrow to different depths in the ground to achieve comfortable microenvironments. Some species are strictly low desert inhabitants whereas others are found only in high desert mountains. When comparing desert rodents to laboratory white rats,

Table 2

Water Turnover Rate per Day in Small Desert Rodents as Compared to Laboratory White Rats and Mice

Species	ml/kg · day	ml/kg$^{0.82}$ · day	Reference
Antelope squirrel	231	523	18
Round-tailed squirrel	117	290	18
Palmers chipmunk	335	721	18
Chisel-toothed kangaroo rat	68	137	18
Desert kangaroo rat	41	92	4
White rat	116	323	4
White mouse	336	585	4

there is no difference except in the case of kangaroo rats. Therefore, it is obvious that kangaroo rats possess special adaptive mechanisms for water metabolism. The economy of water expenditure is not surprising since kangaroo rats depend on metabolic water as the primary source of water intake, whereas other desert rodents utilize food with a higher water content, e.g., greens or cacti. It is interesting to note that the desert kangaroo rat has a slower body water turnover rate than the chisel-toothed kangaroo rat. Food habits and kidney reabsorption may be responsible for the differences between the species. Recently, Mullen [5] reported that water turnover of a desert rodent (Perognathus formosus) in the field ranges from 2.4 to 4.0 times greater than values measured in the laboratory. This is an interesting observation but should be questioned on the basis that his technique, using $D_2{}^{18}O$, is inadequate because the rate of entry of unlabeled water and the rate of re-entry of labeled water from the natural environment were not measured. In his study Mullen [5] showed that T_b in August was 70% longer than that in November. This finding is in disagreement with the known water loss and environmental temperature relationship.

As for large mammals among the desert forms there is a threefold range in water turnover rate, with the camel and the antelope oryx having the slowest turnover rate and the bighorn sheep having the fastest (Table 3). When man, a nondesert mammal, is compared to large desert animals, his water turnover rate falls midway between the two extreme values (Table 3). To further elucidate the concept that phylogenesis is not a major determining factor in water turnover rate, data on large Arctic mammals are presented in Table 3. Although reindeer have a higher value than desert bighorn sheep, musk oxen have a rate of water expenditure almost as low as the oryx or camel.

The turnover rates of energy and water in an ecosystem are known to be biologically linked [6]. Relative to body weight, large animals have a lower metabolic rate than do small ones [7]. When the logarithm of body weight of several species is plotted versus the logarithm of water loss per day, as shown in Figure 2, it is obvious that small mammals have a faster water turnover rate in proportion to body weight. However, the exponential relationship of water to body weight ($W^{0.82}$) differs from that for metabolic rate ($W^{0.75}$). The interspecific corre-

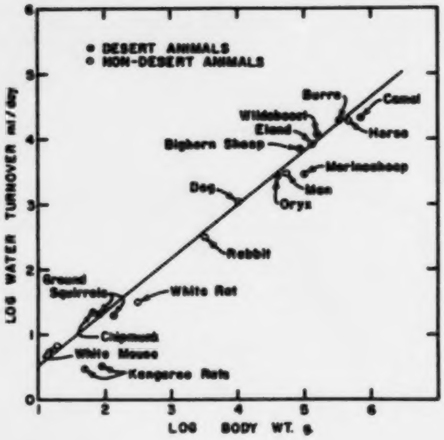
Table 3

Water Turnover Rate per Day in Large Desert and Arctic Mammals as Compared to Man

Species	ml/kg · day	$ml/kg^{0.82}$ day	Referenc
Wildebeest	53	137	11
Eland	78	213	11
Oryx	29	70	11
Camel	29	104	17
Burro	70	190	18
Desert bighorn sheep	86	183	18
Reindeer	128	293	11
Musk oxen	35	99	11
Man	41	87	4

Figure 2

The Relationship Between the Logarithim of Body Weight and Daily Water Turnover Rate



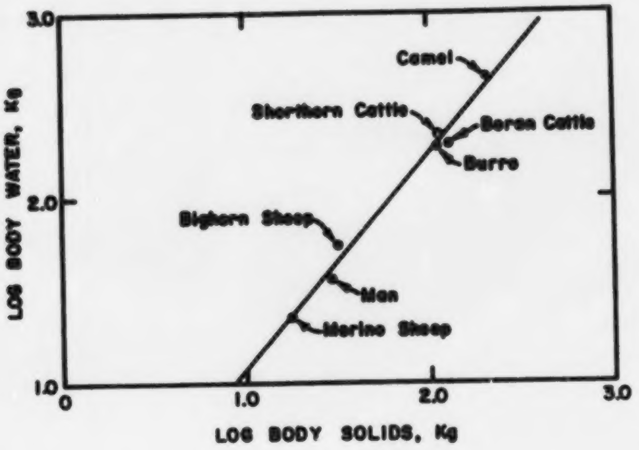
lation between daily water loss and body weight was first shown by Adolph [8] and confirmed by Richmond et al.[4]. It is interesting to see that only kangaroo rats do not fall on the line. This reflects the uniqueness of water metabolism in kangaroo rats and the mechanism by which they maintain their water balance largely on metabolic water. A complete account of water metabolism in kangaroo rats is discussed by Schmidt-Nielson [9].

It would be logical to explain the differences in water turnover rate between species on the basis of their metabolic rates. Although this may hold true in some cases, it does not help in others. For example, kangaroo rats turn over water slowly and also have a 20 to 25% less resting metabolism than the predicted value from the equation which describes the metabolic rate and body weight relationship. However, African antelopes, eland and oryx, have similar metabolic rates [10], but the water turnover rate of the eland is approximately 2.5 times greater than that of the oryx [11]. The difference between species may be explained on the basis of difference in rate of water reabsorption and secretion by the kidney and the intestinal tract.

The question that arises at this point is whether or not tolerance to aridity by some mammals is related to water storage in the body. To elaborate on this question, the logarithm of body water is plotted against the logarithm of body total solids as shown in Figure 3.

Figure 3

The Relationship Between the Logarithim of Total Body Water and Total Body Solids



The ratio of water to solids is constant in all animals studied, a finding reported a few years ago by Richmond et al.[4]. The constant ratio means that the degree of tissue hydration is normal and similar in all animals, therefore the idea of a water depot in the camel or other animals is untenable. The paradox presented by the data in this paper is the fact that while man expends less water than the burro, the burro is superior to man in tolerance to desert life. This is simply because withstanding water shortage is correlated with tolerance to dehydration and conservation of water expenditure. Man cannot tolerate above 12% dehydration without ill effects [12]. However, a 15%

dehydrated burro functions normally and can walk continuously over 30 miles in a 10-hour period without any apparent ill effects [13]. At the end of this walk, the degree of dehydration is about 21%. Another factor which contributes to the success of burros in the desert is their remarkable economy of work performance [14]. They also conserve water by tolerance to hyperthermia [15], excrete relatively dry feces [13], and probably concentrate their urine. The limit for dehydration of burros is not known. The mechanisms underlying tolerance to dehydration have been suggested for the burro [13] and for the camel [9]. The greatest contribution of water loss in both species is from the intra- and extra-cellular fluid volumes and the smallest relative loss is from the plasma volume. Thus the burro and the camel can maintain relatively normal circulation, which is one of the most disturbed functions during dehydration in other animals.

Another example of the puzzling findings in this paper is that the kangaroo rat is more economical in water expenditure than other rodents but less tolerant to high ambient temperature [16]. Survival during the summer months depends on the behavioral adaptation of seeking a comfortable microenvironment during the daytime heat. Its intolerance to heat may be due to dehydration accompanied by hemoconcentration [16].

Water turnover rate is influenced by many factors including, first, the availability of water in the environment, i.e., free water, the partial pressure of water vapor in the burrow and the water content of the food. A second factor is the environmental heat load. Examination of Table 1 indicates that in the burro and bighorn sheep, water turnover rate is faster in the summer than in the spring. A third factor is lactation. A lactating animal expends more water than the nonlactating animal (Table 1). Other factors include age, sex, insulation, and protein and mineral content of food. Available information on these factors are not sufficient to warrant quantitative estimates.

Table 4

Theoretical Calculation of Survival

Species	Body weight kg	Total body water[1]	Dehydration[2] tolerance, %	Water loss 1/day		Survival time/day
				hydration	dehydration[3]	
Camel	600	400	40	18	7	23
Burro	200	130	35	14	5	9
Kangaroo rat	0.100	0.065	10	0.004	0.0016	4

[1] Total body water is assumed to be 65% of body weight in all animals.

[2] Dehydration tolerance as a percent means that the animal can lose that percent of its TBW without any ill effects.

[3] Water loss in dehydrated animals is assumed to be only 40% of the hydration value.

One potentially important line of research which could, if pursued, yield information of biological value would be the study of water turnover rate in dehydrated mammals. By determination of k/day in addition to total body water and the limit for tolerance to dehydration, one can estimate survival time for animals without water. An example of the theoretical calculation of survival times is shown in Table 4. This information is of great value to wildlife management in the desert since the ability of animals to seek food over large areas between widely spaced water holes is a critical factor for thriving in the desert. Also, studies on water turnover rates will help evaluate the water budget of a given ecosystem. Quantitative knowledge of water sources, the population groups, their density and water turnover rate will aid in constructing a model pertaining to water budget of a given ecosystem. In addition to the value of such a model to wildlife conservation and management, it can be used as an index of energy flow in an ecosystem.

In conclusion, water turnover rate differs from one species to another not on the basis of their phylogenetic character but on the basis of efficiency in water expenditure for thermoregulation, and reabsorption and secretion of water by the urinary and digestive systems. Tolerance to dehydration does not depend on the degree of water turnover rate. Daily water turnover rate may be used as an index for estimation of daily water requirements.

ACKNOWLEDGMENT

This work was supported by NSF Grant GB-17126.

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STUDIES ON THE FOOD CHAIN CONTAMINATION BY TRITIUM

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Beyond its numerous industrial and scientific applications, tritium draws attention because it is produced in large amounts by the expanding nuclear industry and the major part is released into the environment as tritiated water. In this form, it will move freely in the biosphere, and it may be ingested by man not only in his drinking water but also in his food. Indeed, where tritiated water is taken up by plants, incorporation of tritium into organic matter will occur during the photosynthetic process. Ingestion of this tritiated material by animals may lead to tritium content higher than expected in organic molecules which are synthesized by an animal.

In order to obtain more information on this subject, several experiments have been carried out since 1968 at the experimental farm of the Center of Nuclear Studies in Mol, with financial support by the Public Health Ministry. Some recent results gained from data of these experiments are presented in this paper.

Materials and Methods

Approximately three hectares (about 7.5 acres) of pastures have been used for the deliberate experimental contamination of grazing land by deposition of tritiated water. The tritiated water was deposited with a motor-sprayer with a 10 m arm length. The experiments on milk contamination resulting from tritiated water or tritiated grass and hay ingestion were performed in a stable, located in Mol, equipped for radionuclide use. The lactating cows produced between 12 and 20 liters of milk daily. Detailed experimental conditions (with the exception of the hay experiment) as well as the analytical methods and radioactivity measurements are described in papers referenced in Table 1.

Table 1

Types of Experiments Performed Since 1968 to Study the Contamination of Grass and Milk by Tritium

	Number of Cows	Activity (THO)			Reference
		Ingested (mCi)	Injected (mCi)	Deposited (μCi/m^2)	
I. INGESTION					
1. Tritiated Water					
a. single oral dose	2	75			2
	1	220			3
b. continuous oral dose (28 days)	2	28(X 1.5 mCi/day)			2
2. Grass, Tritiated Water Applied While Growing In:					
a. spring	3			180	1
b. summer	3			230	1
c. autumn	8			17	1
d. winter - single	3			450	1
- repeated	3			450	1
3. Hay, Repeated Tritiated Water Applications While Growing	2			10 X 2200	This paper
II. INJECTION					
Tritiated Water - Single Intravenous Dose	1		250		3

In the hay experiment, about 250 m^2 of normal pasture were sprayed with 280 liters of tap water containing about 2 mCi of tritium per liter. Spraying was repeated ten times, with a two-day interval between each application. The grass was cut one week after the last spraying and about 1% of the total tritium deposited now remained in this grass. It was transformed into hay by air-drying on the field as is done under normal farming conditions. After drying, the hay was stored in plastic bags.

This hay was later fed to two lactating cows for about one week. Analysis of the hay was performed in the following way: to remove the moisture, a known amount was dried for 24 h at 110°C in an oven. This hay dry matter was submitted to various solvents in the following order: petroleum ether, ethyl alcohol 80%, water, H_2SO_4 (1.25%), NaOH (1.25%).

Results and Discussion

Forage

In the conditions of the hay experiment, the pathways of contamination of plants by tritiated water are multiple. uptake by roots from

Figure 1

Tritium Activity of Milk-H_2O and Milk Fat After Contamination of Pasture at Different Seasons

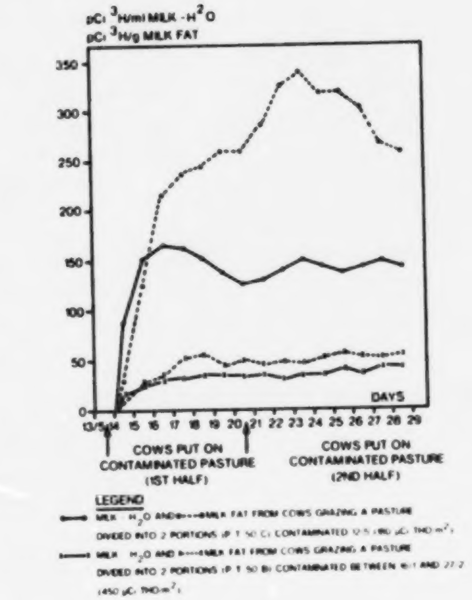

Table 2

Distribution (%) of Tritium in Grass and Hay

Mode of Contamination	Fresh grass (82% moisture)		Air dried grass (13% moisture) (Hay)	
	Water	Dry-matter	Water	Dry-matter
Single deposition (Tritiated water applied eight days before cutting)	65.9	34.1	6.4	93.6
Repeated deposition (Tritiated water applied ten times, two-day intervals). Last application eight days before cutting.	75.5	24.5	8.3	91.7

Figure 2

Evolution of the Grass and Milk Contamination, After a Single Deposition (230 $\mu Ci/m^2$) of Tritiated Water on a Pasture (grass daily cut and fed)

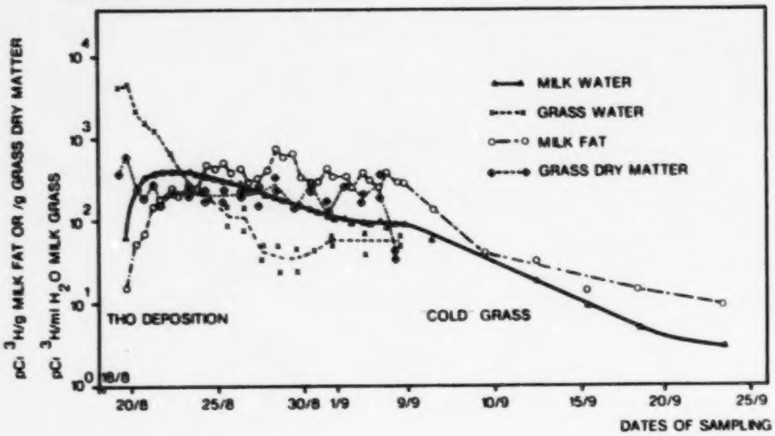

Table 3

Calculated Specific Activities (pCi ^{3}H/gH) of Grass Components After a Tritiated Water Deposition of 1 μCi ^{3}H/gH

	Harvest	Grass	
Mode of Contamination	Days after last deposition	Water (pCi/g H)	Dry-matter (pCi/g H)
Single deposition (0.25 l/m^2)	1 8	2905 216	995 860
Repeated deposition (Ten times 1.1 l/m^2 two-day interval)	8	4655	15520

Table 4

Tritium Contents of Hay* Components

	Extract	
	% initial dry matter**	nCi ^3H/g
Petroleum ether	3.39	475.5
Ethyl alcohol 80%	19.05	14.5
H_2O	6.67	~ 0
H_2SO (1.25%)	45.11	6.9
NaOH (1.25%)		
Residue (cellulose, lignin)	25.78	16.5

*Hay produced from grass contaminated by a repeated deposition (ten times with two-day intervals).

**24 hours drying in oven (110°C).

Table 5

Transfer* "Deposition-to-Milk" of the Tritium Deposited as Tritiated Water on Pastures at Different Seasons

Seasons	Days after deposition	Transfer coefficient to milk components, percent		
		H_2O	Dry matter	Fat
Winter	102	1.2×10^{-3}	1.7×10^{-3}	3.6×10^{-4}
Spring	4	1.0×10^{-1}	1.0×10^{-2}	7.0×10^{-3}
Summer	4	1.6×10^{-1}	-	3.5×10^{-3}

*The rate of transfer is expressed here as the percentage of tritium activity deposited per m^2 which, for a particular milk constituent, is secreted in one liter of milk.

soil, direct foliar uptake, and exchanges of water vapor between the atmosphere and the aerial part of the plants. In the case of a single deposition on a growing pasture [1], the direct contamination of the foliage is the most common route. If one considers the distribution of tritium between the dry matter and the water of forage after a single [1] or repeated deposition, Table 2, one could conclude that this distribution is little influenced by the mode of deposition. However, the "field loss" (or half-life) of the tritiated water of grass is fast (about one day) during the first eight to ten days following the deposition, Figure 2. Moreover, the values of the specific activities, Table 3, clearly show that the level of incorporation of tritium is much higher (15 times) in the case of a repeated deposition than for a single one. For a single deposition, the specific activity of water decreased rapidly, as mentioned above, from the first to the eighth day after the spray. The specific activity of the hay or grass dry matter changed little, however, and the reason for the small decrease observed is likely to be growth of the grass, and not due to catabolic process.

Another point of interest is the labeling of the various components of grass contaminated by a repeated deposition (10 times, with a two-day interval); the tritium contents of these compounds, present in the hay produced by this grass, are given in Table 4. One can see that the fatty fraction, extracted by petroleum ether, is heavily labeled in comparison to other fractions; in contrast, the aqueous extract is practically not tritiated.

These results indicate a heterogenous distribution of tritium specific activity in the grass organic components, contaminated under the experimental conditions. This distribution, therefore, may be reflected in the specific activities of milk components which are formed from precursors in the grass.

Contamination of the milk

A cow's whole milk usually contains between 110 and 130 g of dry matter per liter. The hydrogen content of the main components (g hydrogen/g component) has been determined experimentally (combustion water): 11% in the fat, 7.3% in the casein and 6.9% in the lactose. The contamination of milk produced by cows grazing on pastures contaminated either in winter or in the growing season has been investigated [1]. Data on the transfer, "deposition-to-milk," of applied tritium to milk are given in Table 5. In addition, it is interesting to note the obvious influence of harvest delay for the winter deposition on the contamination of milk produced in spring. This delay effect, on the tritium concentration of water and fat from milk produced by cows eating grass from pastures contaminated at two different times before harvest, is shown in Figure 1.

In order to determine the transfer coefficient, "grass-to-milk," three cows were fed with a known amount of grass contaminated by a single deposition of tritiated water and cut daily starting immediately after deposition. The maximal concentration of tritium in the milk water was observed on the fourth day after deposition; then the content decreased with a half-life of about four days. When, after cessation of the contaminated grass feeding (16 days), the cows were fed with "cold" grass, then the half-life became 3.2 days. These values are similar to those observed in the case of a single tritiated water ingestion [2]. This observation is not surprising because the tritium in the milk water originates almost exclusively from the water of the

Table 6

Transfer* Coefficient "Grass-to-Milk" for Tritium

Days after deposition	Cow No.	Grass**			Milk		Transfer coefficient	
		Amount of grass (kg/day)	µCi ^{3}H/d in tissue water	µCi ^{3}H/d in tissue dry matter	(nCi ^{3}H/g) H2O	(nCi ^{3}H/g) Milk fat	H_2O grass to H_2O milk (%)	Dry-matter grass to milk fat (%)
6	20	70.0	10.3	1.58	331	544	3.26	1.83
	35	71.0	10.5	1.52	357	518	3.16	1.83
	38	71.0	10.5	1.52	330	299	2.24	0.90
16	20	38.5	2.7	1.25	99	287	4.29	0.75
	35	56.0	4.0	1.78	99	220	3.77	0.65
	38	53.0	3.8	1.69	74	268	2.09	0.78

*The transfer coefficient is defined as the percentage of daily ingested tritium which, for the particular milk constituent, is secreted in one liter of milk on the day considered. Delay between ingestion and secretion is one day for water and two days for milk fat.

**Fresh from pasture.

Table 7

Specific Activities (S.A.) of the Milk Constituents After Ingestion of 1 mCi ^{3}H/day by Various Routes of Transfer

Route of transfer	Time elapsed since the beginning of experiment (days)	mCi ^{3}H/g H milk water	S.A. milk fat / S.A. milk water	S.A. lactose / S.A. milk water	S.A. casein / S.A. milk water
Tritiated water ingestion					
a) single oral dose	2	12.8	0.38	0.59	0.53
b) continuous oral dose (daily single dose for 28 days)	28	162.0	0.42	0.29	0.28
Grass (summer) (daily cut and fed to animals)	6	135.0	0.56	--	--
	16	161.0	0.27	--	--
Hay (stored in plastic bags)	7	47.0	16.3	1.8	14.4

grass, which decreases rapidly ("field-loss" or half-life of about one day).

The evolution of milk fat contamination is quite different. The concentration increases rapidly during the first days of feeding, exceeds the tritium concentration of the milk water after about five days, and then remains almost constant during feeding contaminated grass, Figure 2. The transfer coefficient "grass-to-milk" has been computed for each of the three cows from the fourth to the sixteenth day of the feeding experiment; the values of this coefficient at the sixth and the last day are given in Table 6. For determination of the coefficient, a one-day delay between the activity values for grass, milk, and water was utilized; in the case of the coefficient for dry matter and fat, the delay was two days.

In our previous experimental studies [1] on the transfer of tritium from drinking water to the milk dry matter, values of 0.004% and 0.037%, respectively, were found for THO-single oral dose and THO-continuous daily dose during one month. The value of the tritium transfer coefficient from grass to milk organic matter is ten times higher if a cow daily ingests grass contaminated under simulated natural conditions.

Data obtained for the various routes of transfer are given in Table 7 for comparison. It must be borne in mind that the experiments from which these data result are different in a number of aspects [3].

The conclusion from these results is that an increase in ingestion of tritium incorporated in organic compounds, relative to tritium in water form, produces a shift of the activity in milk components from the water fraction towards the organic fractions. It is not clear why lactose should have less tritium incorporation than the other milk constituents during ingestion of tritiated organic compounds. It may be that, during fermentation in the rumen of the cow, most of the tritium is lost in the propionic acid fraction from which glucose (and hence lactose) is synthesized. This hypothesis needs to be verified.

Conclusions

When considering these results as far as their radioecological implications are concerned, it can be said that tritium differs from other radionuclides in that it becomes rapidly distributed in a particular environment when released as tritiated water; at the same time, it is diluted to a considerable extent with light water. This need not be true to the same degree for tritium incorporated in organic material. Thus, the chemical form in which tritium is taken up is an important consideration.

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TRITIUM EXCRETION VIA COW'S MILK AFTER CONTINUOUS INTAKE OF TRITIATED WATER

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According to the recommendations of the ICRP, tritium as water has a half-life of 10 to 12 days [1 - 3]. Recently there have been indications, particularly from persons exposed to higher levels of tritium or for prolonged periods, that the excretion of certain fractions of tritium takes place at a considerably lower rate. In the case of chronic intake, tritium evidently has an excretion rate which is determined by the metabolism of the tritiated tissue rather than by the turnover rate of the body water. It can be estimated that in case of a single intake of tritium, the proportion incorporated into organic matter will not exceed 1% [4 - 6]. Large amounts of tritium administered for a prolonged time result in longer half-lives, which is an indication that the fraction bound to organic matter after chronic intake must be larger than after a single intake [7].

The incorporation of tritium into cow's milk after continuous intake was chosen as a model for our studies on prolonged half-lives because there was a particular interest in two problems. If it can be expected that tritium is excreted in milk not only as tritiated water, but also bound to organic matter, then how does this affect the quantity of tritium taken up by children who might consume tritiated milk? In addition, there was interest in obtaining information on a critical pathway of tritium released to the environment.

Procedure

In the first experiments, a cow was given 100 μCi of tritiated water in a single intake. To obtain major amounts of tritium-labeled milk required for subsequent investigations and to get maximum incorporation into the organic components of milk, two further experiments were run, one for 40 days and one for 150 days. During these experiments, the cow was given a daily dose of 1.5 to 1.7 mCi. On the basis of a daily consumption of approximately 50 liters of drinking water, this results in a concentration of approximately 30 nCi/ml, which corresponds to the maximum permissible concentration in water for occupationally exposed persons. In addition to milk, samples of urine, feces, and blood were repeatedly analyzed for tritium.

To separate the organic components of milk from the aqueous phase, freeze drying (lyophilization) was chosen. A one-liter aliquot of the daily milk production was placed in a refrigerator overnight. The next morning, the sample was separated by centrifugation at about -1°C. From the skimmed milk below the cream layer, aliquots were taken for lyophilization. Fifty ml or more of skimmed milk were frozen in liquid nitrogen by slowly turning the 250 ml flask. A thin layer of frozen milk settled on the inside walls of the flask. Using high vacuum equipment, the free water was removed by reducing the pressure to 10^{-5} torr. Freeze-dried skim milk solids, whose remaining content of free water was 2%, were obtained after four to five hours. To remove the remaining 2% of free water, the solids were washed twice with distilled water and dried in the same manner. The second washing was free of tritium as shown by controls.

In order to measure the tritium bound to organic components of the milk, the lyophilized milk powder was burned according to Pregl in oxygen, using a platinum catalyst and hot copper oxide [8-11]. Combustion-water was absorbed on silicagel (blue gel) and expelled again by distillation with xylene.

Tritium activity was determined by using a liquid scintillation technique. The scintillation solution consisted of 4 g of 2,5-diphenyloxazole (PPO), 0.05 g of (1,4-2,5-phenyloxazolyl)-benzene (POPOP) and 120 g of naphthalene up to 1000 ml with dioxane. A PACKARD-Tricarb liquid scintillation counter was employed, whose background varied between 8 and 22 cpm as a maximum over the whole period of measurements. Standardized solutions were used for calibration; efficiency and stability of background were checked periodically.

Free water in the lyophilized milk powder was determined by titration according to Karl Fischer [12-15]. The same method was employed for testing all reagents for their water content. Appropriate corrections were made.

Figure 1

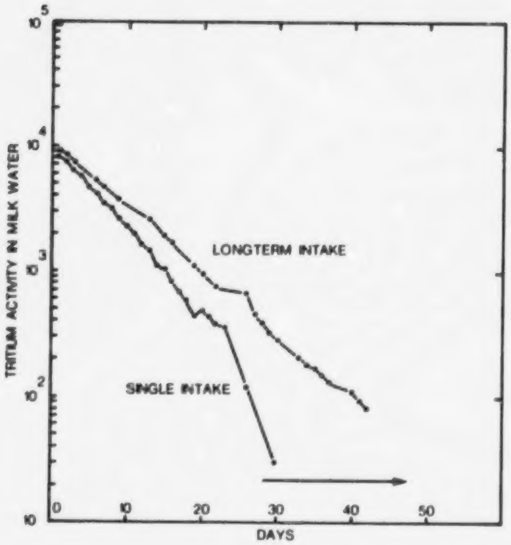


Table 1

Excretion of Tritium After Long-Term Intake

Date	Days after Start	Tritium content of water (nCi/ml) Intake	Milk	Urine	Feces
40 days:					
23,9	6	Ø:32.5 (14/44)	17.4	17.4	-
26,9	9		20.5	19.0	-
3,10	16		26.3	28.8	-
5,10	18		27.2	26.0	-
8,10	21		29.1	28.3	-
27,10	40		38.8	36.4	-
150 days:					
9,2	14	Ø:29.5 (18/40)	27.8	25.1	-
1,6	126		38.8	38.4	38.2
8,6	133		33.0	34.3	33.8

Figure 2

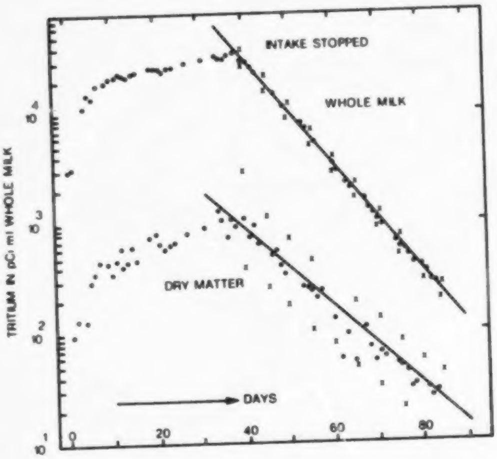


Figure 3

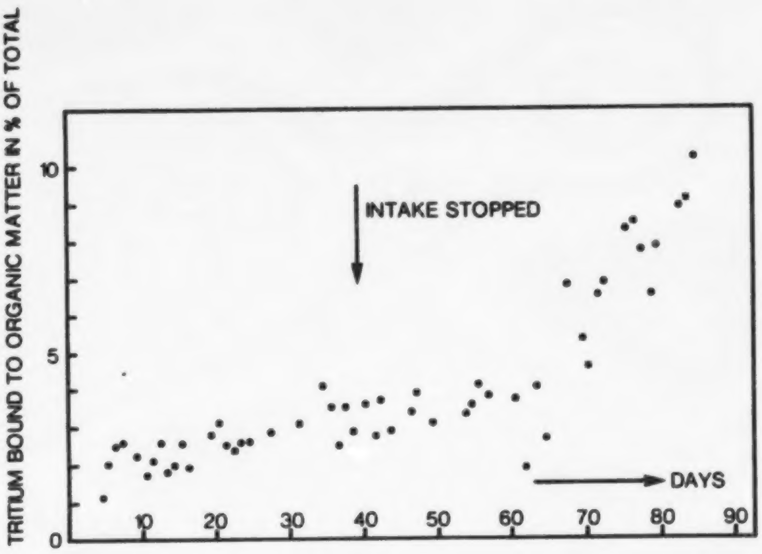
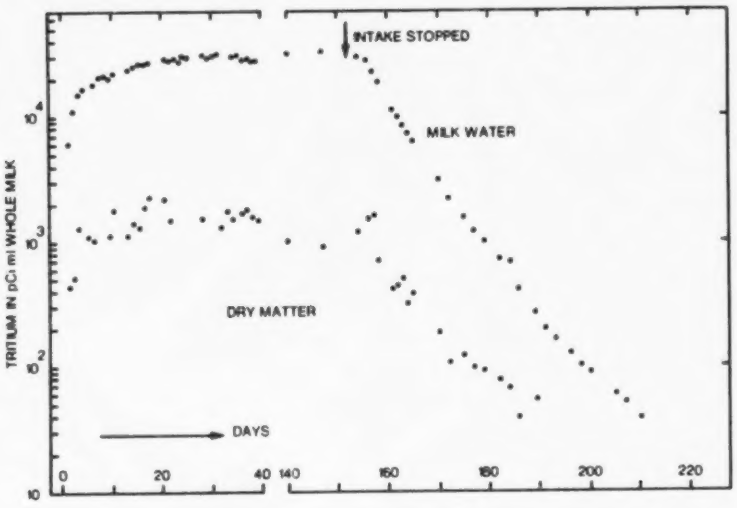


Figure 4



Results and Discussion

After a single intake of 100 µCi of tritiated water, tritium was excreted with a half-life of about four days, as shown in Figure 1. This corresponds to excretion rates via urine which have been established by others [16].

After an intake of a daily quantity of 1.5 to 1.7 mCi of tritiated water over a period of 40 days, it becomes evident that tritium is excreted in about equal concentrations in milk, water, urine, and in water contained in feces, Table 1. After some 16 to 20 days, an equilibrium is established between the intake via the GI-tract and the excretion via milk or urine. The concentrations of tritium in the water of the milk and that of urine are equal within biological variations, Figure 2.

After the 40-day intake, the activity present in the aqueous phase decreases more rapidly than that in the solid portion of milk; the proportion of tritium present in organic matter increases steadily. At the end of ingestion, it is about 5% percent of the total present in the whole milk. After another 40 days, the tritium content reaches some 10% of the total excreted with whole milk, Figure 3.

Statistical analysis of corresponding sections of the two curves indicates that the slopes vary significantly and are thus an indication of tritium binding to organic matter. The calculated half-lives for these curves are five days for the whole milk and seven days for the dry matter, Figure 2.

Table 2

Specific Activity of Milk Water and Combustion Water of Solids and Fat of Cow's Milk (150 days)

		Tritium content of water (nCi/ml)				
Date	Days after Start	Milk	Solids	Ratio	Fat	Ratio
1,6	126	38.8	8.0	4.9	7.6	5.1
8,6	133	33.0	9.7	3.4	5.3	6.2
15,6	140	38.0	8.1	4.7	4.8	7.9
22,6	147	39.0	7.3	5.3	6.4	6.1
29,6	154	34.9	9.6	3.6	7.1	4.9

As shown in Table 2, the specific activity (pCi per mole H) of combustion water is different from the specific activity in the free water of the milk. When comparing the curves for milk water and dry matter, one might assume excretion after chronic intake to be described better by a multiexponential function than by a single exponential curve.

The long-term experiment over 150 days showed that tritium bound to the solid mass of milk was again 5 to 10%, relative to total tritium in the whole milk. Thus this fraction is of the same order as the fraction of dry matter in milk itself. Milk is made up of approximately 87% water and 12 to 13% dry mass. Of the dry matter, approximately 3.5% is fat and 9% fat-free solids. These latter are composed of 3.6% protein, 4.7% lactose, 0.7% mineral salts, and 0.025% N not bound to protein.

In the 150-day sequence, a little shift can be noted towards longer half-lives, and the curve is no more a straight line. But the excretion rate does not change to such a degree that the overall exposure will be much greater than after single intake. The overall half-life does not change considerably in comparison to that of the 40-day intake.

An explanation of this phenomenon is given by recent findings on lactation. Lactation of a cow is characterized by a high turnover of matter. Within a period of 24 hours, 20 to 30 kg or more of milk are produced. The organic components are synthesized in that time period. Milk is not the reflection of the blood proteins which, after some slight transformation, are formed into milk proteins; it is synthesized from the simple components present in the blood pool, such as amino acids, etc., by the epithelium of the mammary gland, which each day forms the macromolecules of casein, lactalbumin and lactoglobulin, the triglycerides of milk fat, and lactose [17,18]. Despite the high turnover rate, there is no doubt that milk proteins and their metabolic products may be tritiated, if an increased level of tritium is maintained within the cow's body water.

If there is an equilibrium between the intake of tritium from the environment and the animal's excretion, the fraction of tritium bound to organic matter in the excreted milk is between 5 and 10% of the total present in milk. This means that tritiated compounds are directly introduced into the metabolism of men.

Milk fat, too, contains tritium bound to organic matter. The combustion water of milk fat (thoroughly washed) and dry matter free of fat and water contain about the same quantity of tritium. After feeding tritium water to a level corresponding to the MPC_{water} of ICRP, the specific activity in milk fat is 8.4 μCi/mole. If we say this is all glycero-tripalmitic acid, we have a hydrogen-tritium ratio of about $1:10^{-12}$, as compared to a natural tritium content of the atmosphere of $1:10^{-18}$.

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BIOLOGICAL IMPLICATIONS OF THE APPLICATION OF TRITIATED LUMINOUS COMPOUNDS

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The production and application of tritiated luminous compounds are two endeavors in which the industrial worker may be exposed to tritium. These paints or compounds also represent a potential hazard due to the possibility of absorption, inhalation, or even ingestion in the case of small children, of the luminous compounds or released tritium. It is, therefore, necessary to acquire a better understanding of the biological characteristics of the tritium released from this source.

Various aspects of tritiated luminous compounds have been studied, including their tritium release [1-3] and biological significance [4-9]. To the present time, the following luminous compounds have been investigated:

1. An aliphatic acid with a specific activity of 200 mCi/g, manufactured by 3M Company and herein abbreviated as MH 200.
2. A polystyrene compound with a specific activity of 250 mCi/g, available from Conrad Precision Company and herein abbreviated as CH 250.
3. A silicone rubber with a specific activity of 250 mCi/g, available from Radium Chemical Company and herein abbreviated as RH 250.
4. The same compound with a specific activity of 20 mCi/g, herein abbreviated as RH 20.
5. A polystyrene compound with a specific activity of 600 mCi/g, produced by New England Nuclear Corporation and herein abbreviated as NH 600.
6. A polyester compound, studied by Remy et al. [11], with a specific activity of 65 mCi/g, herein abbreviated as RC 65.

Tritium Absorption

It can be seen from Table 1 that the cutaneous absorption of tritium from luminous compounds is dependent upon the chemical nature of the compound and the decontamination procedure following exposure. Rehnberg et al. [7], have demonstrated that the most practical and efficient manner for decontaminating exposed skin areas is to wash the exposed area with warm water, monitoring with UV light to ensure the removal of all possible luminous particles. This procedure results in a 20-fold reduction in tritium absorption, Figure 1. The chemical nature of the luminous compound is the predominant factor in determining the percentage of absorbed tritium. The more biologically active the compound, the greater the tritium absorption. Compound MH 200, an aliphatic acid, under identical conditions resulted in cutaneously absorbed tritium levels 88 times higher than that of compound NH 600, a

Table 1

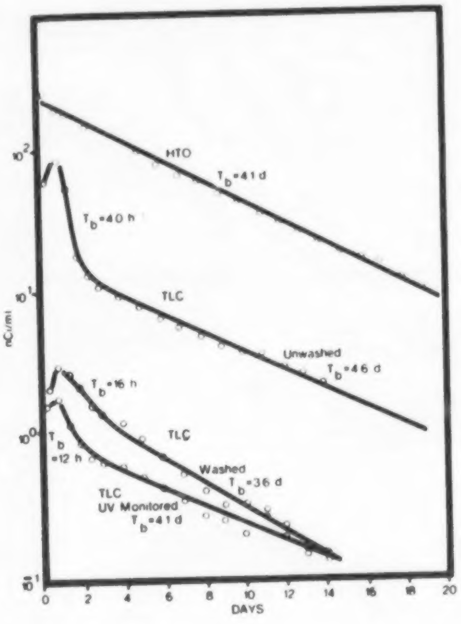
Comparison of Absorbed Tritium Activity Through the Skin of Rats from Various Luminous Compounds

Compound	Contact Time (h)	% Absorbed	Ts (d)	Tw (d)	Remarks	Ref.
MH 200	8	8.83	0.5	4.2	Washed and UV monitoring*	10
CH 250	8	0.59	0.4	3.5	Washed and UV monitoring*	10
RH 250	8	0.19	0.5	3.2	Washed and UV monitoring*	10
RH 20	24	0.14	0.5	4.1	Washed and UV monitoring*	7
RH 20	24	0.24	0.7	3.6	Skin washed with water only	7
RH 20	24	2.4	0.2	4.6	Skin unwashed	7
NH 600	8	0.10	0.3	3.2	Skin washed and UV monitoring*	10
HTO Control		100.0		4.1		7

*Area washed with warm water and monitored with UV light to ensure removal of all possible luminous particles.

Figure 1

Comparison of the Biological Half-lives of Tritium in Rats Absorbed via the Skin Following Various Decontamination Procedures from Luminous Compounds and HTO Injected Control Animals [7]



polystyrene compound, and 63 times higher than RH 250 and RH 20, which are silicone rubber based compounds. It has been demonstrated that the absorbed tritium from compounds MH 200 and RH 20 can manifest chromosomal aberration in kidney and lung cells after eight hours contact time [9]; care, however, is advised in interpreting these chromosomal aberrations due to the questionable nature of the experimental controls.

The absorption of tritium from luminous compounds via the GI tract has also been investigated and is summarized in Table 2. The fraction of tritium absorbed by the GI tract is dependent upon the chemical nature of the luminous compound and the species of experimental animal. Twenty-one percent of the tritium in the biologically active aliphatic acid compound, MH 200, was absorbed by the cat's GI tract while less than one percent (0.7 percent) of the tritium in the polystyrene compound, NH 600, was absorbed [10]. Remy *et al*. [11] found that 80 percent of the tritium from a tritiated polyesteric compound, RC 65, was absorbed by the rat's GI tract. This high absorption of tritium can partially be explained by the more desiccating nature of the rat's digestive system than that of the cat's. Wheeler *et al*. [12], found that the rat's GI tract absorbed six times more tritium from a particular luminous compound than did the cat's GI tract.

Table 2

Summary of Tritium Absorption and Elimination via the GI Tract

Compound	% Absorbed	Ts (d)	Tw (d)	Animal	Reference
MH 200	21.4	0.6	9.8	Cat	10
RH 20	3.0	1.0	7.8	Cat	12
CH 250	4.2	0.4	7.7	Cat	10
RH 250	2.7	0.4	8.8	Cat	10
NH 600	0.7	0.6	8.3	Cat	10
RC 65	80.0	1.2		Rat	
RH 20	19.0		4.6	Rat	12
HTO Control	100.0		4.1	Rat	12
HTO Control	100.0		7.4	Cat	10
HTO Control	100.0		8.4	Cat	12

Tritium Elimination

Absorbed tritium from luminous compounds, regardless of route of absorption, is eliminated by at least two distinct half-lives, Figures 1 to 3. The first half-life, Ts, resembles that of biologically foreign materials while the second half-life, Tw, resembles that of tritiated water.

It has been demonstrated that the tritium eliminated during the Ts is not entirely aqueous in nature; in one experiment, 81 percent of the tritium eliminated 24 hours post-administration was nonaqueous in nature while 100 percent of the tritium eliminated eight days post-administration was aqueous in form [10]. The three separate T1 groups (cutaneous absorbed, GI absorbed, and tritiated water injected) were tested for significant differences in means. The F ratio, based on a

Figure 2

Comparison of the Biological Half-lives of Tritium Absorbed via the G.I. Tract from Luminous Compounds and HTO Intraperitoneal Injections in Cats [13]

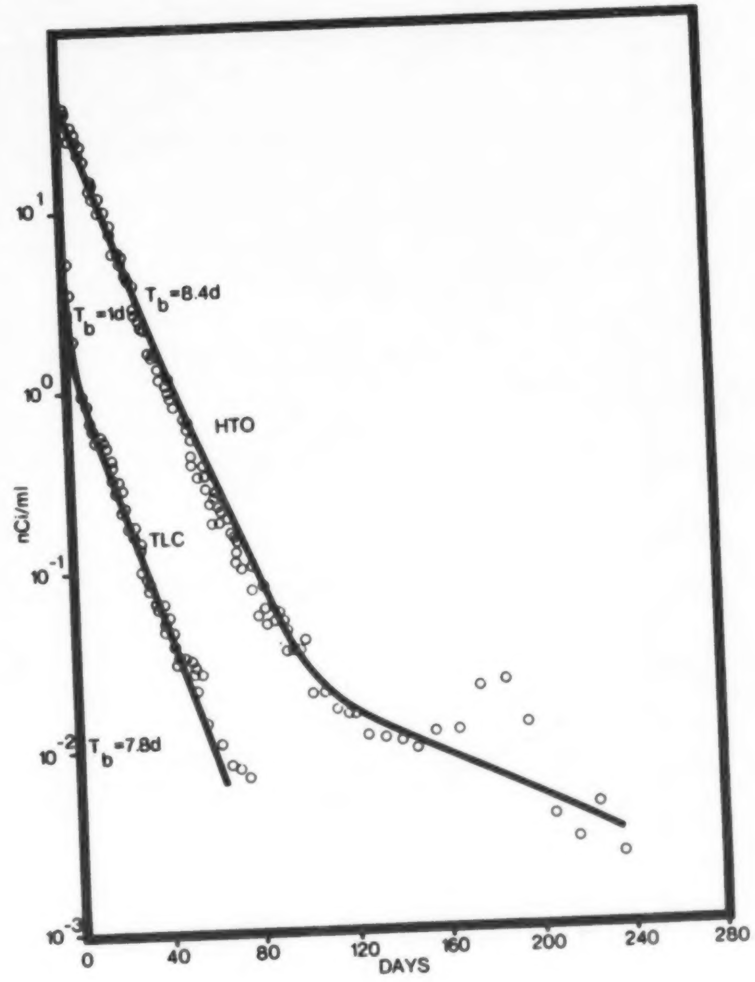
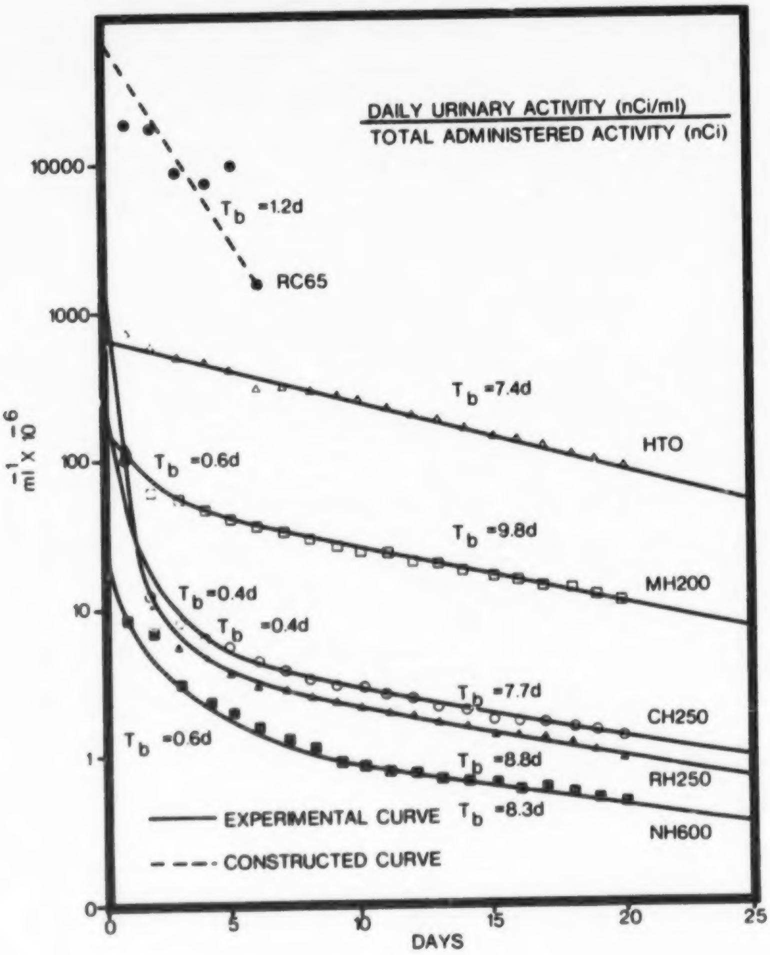


Figure 3

Comparison of G.I. Absorbed Tritium Activity by the Cat



two-tailed F test, was significant at the 95 percent significant level; hence the equality hypothesis was rejected. Remy's value of 1.2 days, the single observation for rat's GI absorption of luminous compounds, was found to be significantly different from the other means. Therefore, the two groups, which had no significant difference in their means and which had a sample size of at least five, were pooled, and an estimate of the mean or Ts with 95 percent confidence limit is given as 0.53±0.2 days. This half-life can be attributed to a large amount of nonaqueous or nonabsorbed tritium which is being eliminated in a manner similar to that of biologically foreign material. The absorbed tritium eliminated under the dominance of Ts is dependent upon route of absorption and chemical nature of the luminous compound. Substantially more tritium is eliminated during the dominance of Ts from experimental animals dosed through the GI tract as compared to those dosed cutaneously, Table 3. Eighty-nine percent of the GI absorbed tritium from RH 250 was eliminated while only 23 percent of the cutaneously absorbed tritium from MH 200 was eliminated under the dominance of Ts [11].

The second biological half-life is independent of chemical form, but dependent upon route of absorption and experimental species, Tables 1 and 2. The three separate T2 groups (cutaneous absorbed, GI absorbed, and tritiated water injected) were subjected to an analysis of variance to determine if there was a significant difference in the means. There was no significant difference at the 95 percent significant level. However, Remy's Tw data of 14 days is significant at the 92 percent significant level. It seems unlikely that the tritium absorbed from a polyester source would have a half-life approximately 3.5 times longer than the tritium absorbed from a totally absorbable source, namely tritiated water. Due to Remy's sampling schedule (1, 2, 3, 4, 5, 6, and 25 days), it is likely that he missed the body of a true Tw, and that his 25th-day sample actually reflects the start of what has been demonstrated to be a third biological half-life [14,15]. If Remy's Tw of 14 days is excluded and the remaining data pooled,

Table 3

Percent Total Urinary Tritium Elimination Due to Various Components

	Ts			Tw		
Compound	Cutaneous Absorbed	GI Absorbed	Ratio*	Cutaneous Absorbed	GI Absorbed	Ratio*
MH 200	23.1	23.5	1.0	76.9	76.9	1.0
CH 250	40.3	79.9	2.0	59.7	20.1	0.3
RH 20	43.1	88.6	2.0	56.9	11.4	0.2
RH 250	43.1	88.6	2.0	56.9	11.4	0.2
NH 600	41.3	58.7	1.4	58.7	44.9	0.8
RC 65		80.0			20.0	

*Ratio of GI to cutaneous

statistical analysis reveals no significant differences in the means at the 50 percent significant level. Therefore, it may be concluded that Tw for tritium absorbed from luminous compounds by the rat's GI tract is the same as the Tw for tritiated water injected control rats and is given with 95 percent confidence limits as 3.9±0.4 days.

There is no significant difference in the Tw for tritium absorbed from tritiated water or from luminous compounds by the cat's GI tract. The probability of the absolute value of T statistic exceeding 0.8348 is 20 percent. Therefore, the two sets of data were pooled, and with 95 percent confidence limits, the overall mean or Tw is given as 8.3±0.7 days. With one exception, a larger fraction of cutaneously absorbed tritium is eliminated under the dominance of the Tw than Ts. This exception is compound MH 200 which is also the only compound exhibiting elimination independent of absorption route, Table 3.

Conclusion

The amount of tritium absorbed from luminous compounds depends on the route of absorption, the chemical nature of the luminous compound and the test species. The rat is the only animal that has been investigated for both cutaneous and GI tract absorption of tritium from luminous compounds. Wheeler *et al*. [12], found that 19±4% of the tritium was absorbed through the gut while Rehnberg *et al*. [7], found only 0.14 to 2.4% of the same compound was absorbed through the skin depending on decontamination procedures.

Tritium absorbed from luminous compounds is eliminated by at least two distinct half-lives; the first half-life, Ts, is composed of primarily nonaqueous or nonabsorbed tritium while the second half-life, Tw, corresponds to aqueous tritium. The close agreement between the controls and the second experimental biological half-life indicates that the tritium absorbed from luminous compounds is handled and eliminated in a manner similar to body water. The absorption is to a great extent based on hydrogen exchange and metabolic reactions leading to the formation of metabolic water. The fraction of absorbed tritium eliminated during each half-life is determined by chemical form of the luminous compound and the route of absorption of the tritium. Tritium absorbed via the GI tract, with the exception of tritium from compound MH 200, is eliminated from the body faster than cutaneously absorbed tritium. The biological kinetics of tritium absorbed from luminous compounds is similar to that of tritium from any other source.

NOTE

Use of a brand name in this report does not imply endorsement by the Environmental Protection Agency.

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TRITIATION OF AQUATIC ANIMALS IN AN EXPERIMENTAL MARINE POOL

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In the future, increasing amounts of tritium may be released in the hydrosphere if the power needs of our increasing world population are met with nuclear energy. Currently, 21 nuclear power plants are in operation in the United States, more than 50 are under construction, many are in planning stages, and further expansions are under consideration [1]. Because of the need for coolant water, many reactors are located near inland and coastal waterways. Consequently, it can be expected that in the future expansions of the nuclear power industry, tritium may be added to estuarine and offshore ecosystems that have been and still are important sources of food for man.

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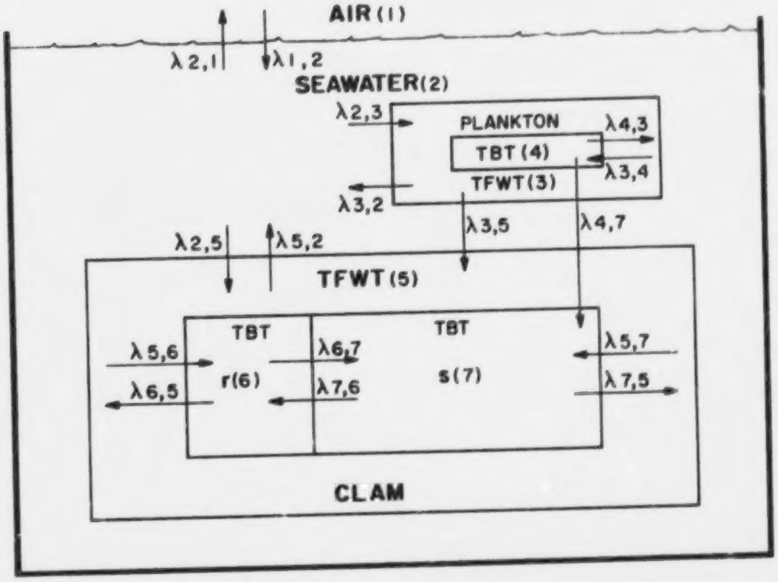
Relatively little information is available on the rates of tritiation of tissue free water and organic matter of aquatic animals. Some data on tritium in freshwater animals were obtained by Skauen [2] who raised guppies in tritiated water to determine whether they exhibited any toxic effects. He found that about 90% of the tritium in the fish was in the body water and only 10% was incorporated in the body tissues. He states the tritium in the fish equilibrated with that in the surrounding water but presents no data on the rates of tritiation of the hydrogen compartments or on their final specific activities. Data on marine animals are restricted to a few measurements by Koranda [3] on the levels of tritium in the loose and bound water of marine organisms sampled from nuclear craters in the Pacific Proving Ground. Koranda states that animals from some of the areas had "an enriched tissue-bound tritium content."

Figure 1



In aquatic animals placed in a tritiated ecosystem, tritium in the water and in their food would be expected to equilibrate with the hydrogen compartments. The incorporation and loss of tritium in an animal could then be described by a model. A simplified scheme of the expected movement of tritium in clams is given in Figure 1. The two main compartments in the animals are the hydrogen in the body water and the hydrogen in the organic material. The tritium in the body water compartment is referred to here as the tissue-free-water tritium and that in the organic material as tissue-bound tritium.

The tissue-bound tritium is usually considered to have a minimum of two subcompartments, the tritium bound to oxygen, nitrogen, and sulphur being in a compartment r that is rapidly exchangeable, and that bound to carbon in a compartment s that is very slowly exchangeable. The main routes of entry into the tissue-bound-tritium pool are from the tissue-free-water tritium of the clams and the tissue-bound tritium of their food; the main routes into the tissue-free-water tritium are from the ecosystem water and the tissue-free-water tritium of the food.

An investigation was initiated to obtain experimental data on the rates of incorporation and loss of tritium in the tissue-free-water tritium and tissue-bound tritium pools of marine animals, so that the model could be adapted for predictive purposes. Tritiated water was added to an ecosystem containing clams, crabs, and fishes; then the changes in tritium concentration with time were followed in the water and in the tissue-free-water tritium and tissue-bound tritium of the animals. The tritiated water was added to the ecosystem as a single injection to simulate an accidental release from a nuclear reactor or a nuclear detonation in or near an aquatic ecosystem.

Materials and Methods

A fiberglass pool 10 feet in diameter and 3 1/2 feet deep, with a substrate of poorly sorted sediment, was filled with 2000 gallons of seawater. The water in the pool was circulated continuously by two pumps, each with a flow rate of about nine gallons per minute. The water intakes of the pumps were near the bottom center of the pool whereas the outflows were at the surface on the periphery. Air was distributed to the bottom of the pool from four gas-dispersion tubes spaced at equal intervals around the periphery. The temperature of the water was recorded daily.

The seawater added to the pool was obtained from the Steinhart Aquarium, San Francisco, CA; it was obtained by pumping from a pipeline that extends 3 1/2 miles into the ocean [4]. The water was filtered at the Aquarium, then transported to Lawrence Livermore Laboratory in a 2000-gallon fiberglass tank obtained and used exclusively for transporting seawater.

The pool was stocked initially with several hundred clams (Mya arenaria), about 50 crabs (Cancer productus), and 50 fishes (Gillichthys mirabilis). All the experimental animals were collected from San Francisco Bay, CA. Then tritiated water was added to a concentration of 77 nCi/ml. Water volume was maintained by the addition of distilled water to replace that lost by evaporation. This maintained the specific gravity of the water at about 1.018, its original value. The animals were given no food during the experimental period. However, algae and detritius present in the water could have been eaten by the clams. The animals were checked daily and dead animals were discarded.

The water and organisms were sampled at intervals of one to three weeks. At each sampling, four clams, four fishes, one or two crabs, and triplicate samples of the water were removed. The water was lyophilized (vacuum distilled) and then analyzed for tritium by liquid scintillation. The animals were dissected, and the like tissues and organs from each kind of animal were pooled to give an "averaged" sample.

The clams were prepared for analysis by removing the soft tissue (typically 30 to 50 g each) from the shells and dissecting the following parts: mantle including the muscular edge, gills and labial palps, visceral mass, muscle (shell adductors, siphon, and foot), body fluid, "peel" (the nonliving material on the exterior of the siphon and mantle edge).

The fishes (30 to 50 g each) were killed; their heads, tails, fins, and viscera were removed, and the lateral muscle masses were separated from the skin and bones. The following parts were prepared for analysis: liver, other viscera, muscle mass, and bones.

The crabs were cooled on ice, their appendages and carapaces were removed, and then their soft parts (50 to 90 g) were dissected. The following parts were analyzed: viscera, gills, and muscle.

For each sampling period, the tissues from each kind of animal were pooled and then lyophilized for 48 to 72 hours to remove the tissue-free-water tritium. The dry tissues resulting from lyophilization were pulverized separately in a mixer/mill by shaking with a Teflon ball in a thick-walled plastic container and then dried to constant weight in an oven at approximately 100°C. In our oven, which had no provision for removing moisture from the circulating air, the loosely bound exchangeable tritium from the dried tissues was almost completely removed, so that the tritium remaining in the tissues after heating represented primarily the slowly exchangeable tissue-bound tritium. Pulverized samples (50 to 200 mg) of the dry material were burned in triplicate in a Packard Tritium Oxidizer to obtain the water of combustion. The hydrogen content of the tissues was determined with a Carbon, Hydrogen, Nitrogen Analyzer (F and M Scientific Division of Hewlett Packard).

The rate of loss of tritium from a group of clams was also determined. Approximately 130 clams were transferred to the University of California Bodega Marine Laboratory after 68 days of tritium accumulation in the pool, during which the tritium of the water fell from 40 to 27 nCi/ml. The animals were held there in aquaria supplied with unfiltered seawater pumped directly from the ocean.

Results and Discussion

Tritium in the Pool Water

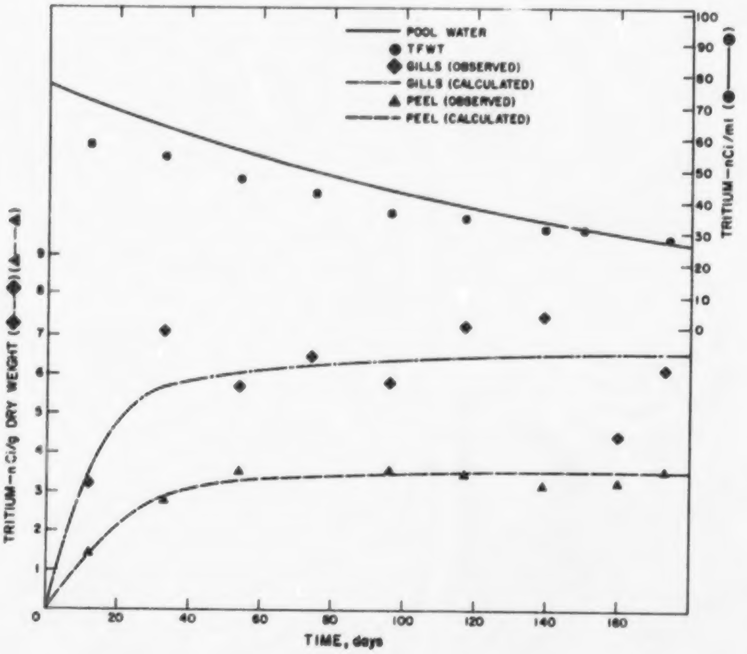
The concentration of tritiated water in the pool decreased with time due to replacement of the evaporated water with distilled water. If the rate of evaporation of water was constant, the concentration should decrease exponentially. A plot of the logarithm of the concentration vs. time gave a straight line with a slope of -0.0063/day; this is the evaporation rate constant ($\lambda_{2,1}$ in Figure 1) used in the calculations of accumulation rate in the clams. A regression analysis of the data gave a correlation coefficient of -0.99. The half-time of tritium in the pool water was 110 days.

The experiment was initiated in winter (December) and was terminated the following summer (June). Since there were no temperature-regulating devices on the pool, the temperature of the pool was dependent upon the air temperature. The weekly mean temperature of the pool water ranged from 7 to 18°C. The different species of animals were exposed to different temperature extremes due to different residence times in the pool water; the ranges (7 to 13°C for the crabs, 7 to 14°C for the fishes, and 7 to 18°C for the clams) were within the natural tolerance ranges of the several species.

Tritium Uptake in Clams

The tissue-free-water tritium of the clams increased to about 95% of the level of the pool-water tritium and then followed the decrease in pool-water tritium during the remainder of the experiment, see Figure 2, indicating rapid exchange between the tissue water and pool water. Since the tissue-free-water tritium of the animals and the

Figure 2



tritium concentration of the pool water were similar, the tissue-free-water tritium also decreased exponentially.

Among the tissues of the clam, the gills contained the highest levels of organically bound tritium, approximately 6 nCi/g dry weight after the first month of exposure and then increasing gradually, (Figure 2). In the nonliving "peel," the concentrations after the first month varied around a value of 3.5 nCi/g dry weight, (Figure 2).

The second highest tissue-bound tritium levels were found in the viscera, where the maximum level was about 5.5 nCi/g,(Figure 3). lowest concentrations (about 2.5 nCi/g) were found in the mantle and muscle; their similarity in concentration was probably due to the high degree of muscularization of the mantle edge.

The extent of tritiation of the organic molecules of the clam tissues can be approximated by comparing the specific activities of the tissues to that of the tissue free water and the pool water. For muscle the hydrogen content was determined analytically; for the other tissues it was approximated from our data on the percentage of organic material, Table 1, from data of Giese [5] on the biochemical composition of bivalve molluscs, and from data compiled on the hydrogen content of carbohydrates, proteins, and lipids [6]. The percentage values for tissue hydrogen used in the specific activity calculations were 6.8 for muscle and mantle, 6.5 for viscera, 7.0 for gills, and 3.2 for "peel."

Figure 3

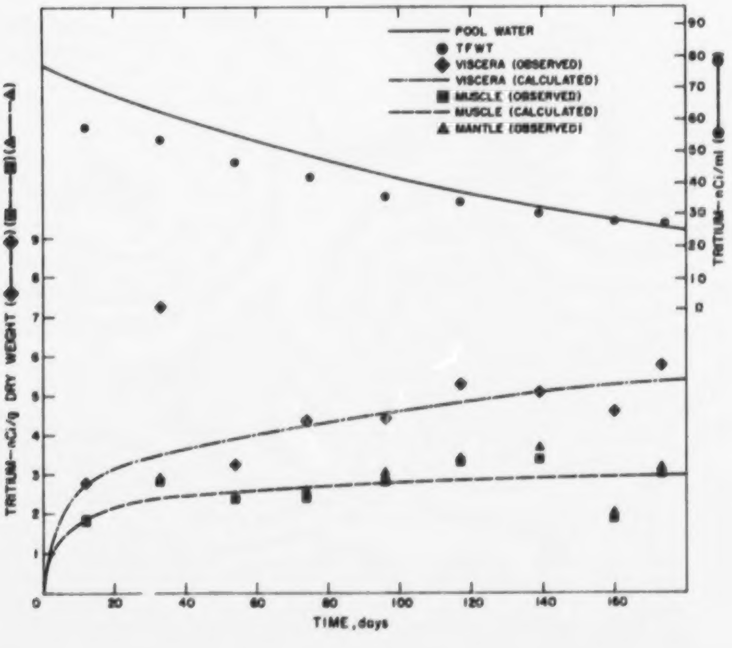
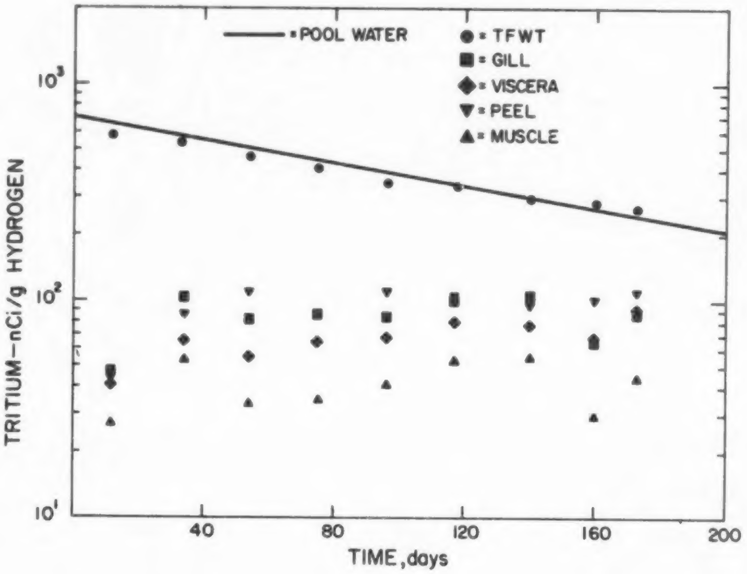


Table 1

Measured Percentages of Organic Material
Per Dry Weight in Clam Tissues

	Organic material (% total dry wt)
Mantle + muscle	93.0
Gills	88.8
Viscera	89.9
Peel	46.3

Figure 4



In the clams, the specific activity of the tissue-free-water tritium rapidly approached that of the pool water and remained at about that level. The specific activity of the tissue-bound tritium changed with time and at different rates for most of the tissues and did not reach that of the water for any tissue, see Figure 4.

The ratio of the specific activity of the tissue-bound tritium to that of the tissue-free-water tritium is frequently used as an index of the relative degree of tritiation of the tissues. In the clams, at the end of the experiment the ratio was highest in the "peel," about 0.5, and lowest 0.2, in the muscle. However, it is shown below that when the concentration of the tritium in the water is not constant, the value of this ratio is determined by the turnover of water in the ecosystem and the turnover of tritium in the tissue. Consequently, it is of limited usefulness in the interpretation of steady-state concentration factors.

Figure 5

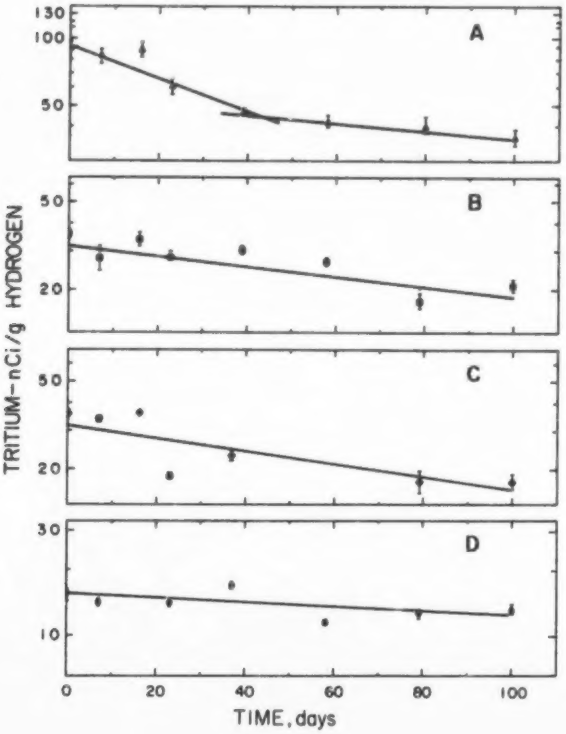
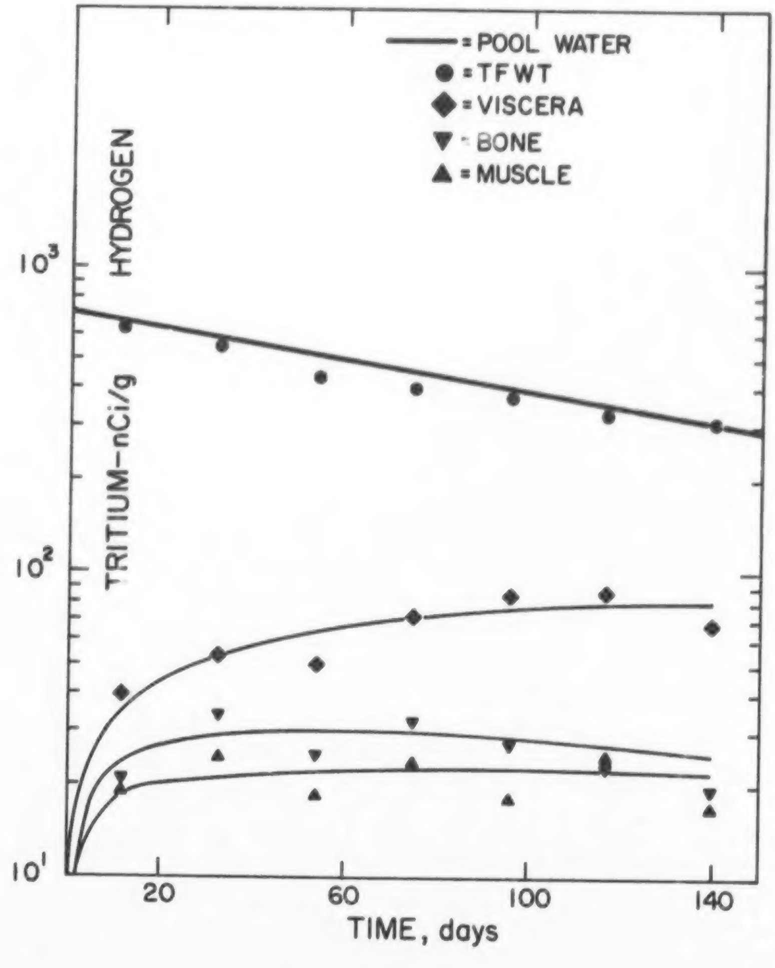
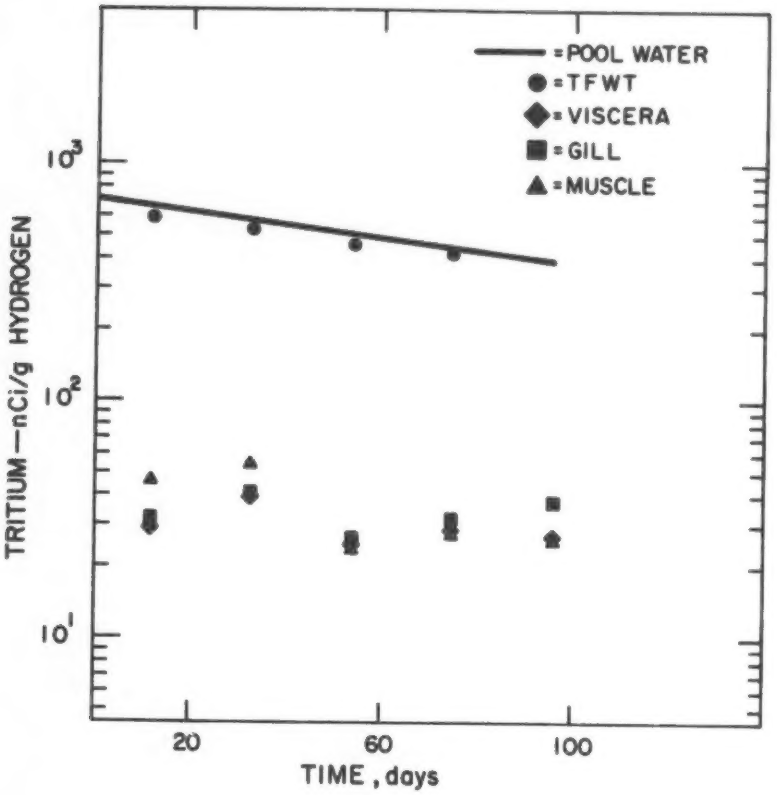


Figure 6



= POOL WATER
● = TFWT
◆ = VISCERA
▼ = BONE
▲ = MUSCLE
HYDROGEN
TRITIUM-nCi/g
10^3
10^2
10^1
20
60
100
140
TIME, days

Figure 7



Tritium Loss in Clams

After 68 days of exposure to tritiated water, the clams were transferred to untritiated, unfiltered, continuously flowing seawater at the University of California Bodega Marine Laboratory. This water is relatively low in environmental pollutants and contains organisms upon which the animals normally feed. The tissue-free-water tritium of these animals decreased very rapidly to essentially background levels. The exchangeable fraction of the tissue-bound tritium was considered to contain little tritium since experiments in progress indicate that it is also rapidly turned over in the clam. Semi-log plots of tissue-bound tritium vs. time were made and the slopes of the lines were determined by linear regression analysis, see Figure 5. A biological half-time (T_b) of approximately 290 days was calculated for muscle, 190 days for "peel" (long-lived component), 150 days for gills and 110 days for viscera.

Tritium Uptake in Fishes

As in the clam, the tissue-free-water tritium of the fishes quickly reached the levels of the pool water and decreased at about the same rate, see Figure 6; the data are expressed as nCi/g hydrogen. The tissue-bound tritium in the viscera reached a maximum of about 90 nCi/g hydrogen after 100 days of exposure. At that time, the tissue-free-water tritium was 340 nCi/g hydrogen. Bone and muscle reached a lower maximum of 31 nCi/g hydrogen after 30 days exposure. These values were calculated from measured hydrogen concentrations in fish muscle where analysis gave 6.8% hydrogen. Values for viscera and bone of 6.3% and 5.0%, respectively, were approximated from data on percentages of organic material in the tissues.

Tritium Uptake in Crabs

As in the other organisms, the tissue-free-water tritium closely followed the pool water levels at all the times sampled, see Figure 7. Tissue levels varied between 22 and 45 nCi/g hydrogen; these levels were reached rapidly. More data are needed before conclusions can be drawn on the turnover of hydrogen in crab tissues.

Mathematical Model

The concentration of tritium in an animal is dependent on its concentration in the ecosystem and on the rate of its accumulation and loss by the tissues. Let us consider the model for clams presented in Figure 1. Our mathematical treatment of the kinetics of accumulation and loss in clams can be simplified considerably by assuming instantaneous equilibration between the tissue-free-water tritium of the clams and the ecosystem water, and between plankton and the ecosystem water. This assumption will not affect the results significantly, since the turnover of tritium in these compartments is very rapid compared to that in the tissue-bound tritium.

The change in tissue-bound tritium concentration in a clam tissue with time, assuming first-order kinetics, can be described by

$$\frac{d(T)}{dt} = \lambda W(t) - \lambda' T(t), \tag{1}$$

where W(t) is the concentration of tritium in the ecosystem (water + food) in nCi/g hydrogen at time $\underline{t}$, T(t) is the concentration of tissue-bound tritium in the tissue in nCi/g hydrogen, λ is the biological accumulation rate constant, and λ' is the biological loss rate constant. When steady-state conditions of uptake and loss have been reached, $\left[d(T)\right]/dt$ = zero and $\lambda W(t) = \lambda' T(t)$. If it is assumed that there is little or no difference in the behavior of tritium and hydrogen, i.e., that the concentration factor for tritium is approximately one, then under steady-state conditions $\lambda \cong \lambda'$. The concentration factor is defined as the concentration of tritium per gram hydrogen in the tissue divided by the concentration of tritium per gram hydrogen in the water.

Tritium can be released into an ecosystem at a relatively constant rate or in a single pulse. Under the first condition, the concentration in the water remains relatively the same with time at a single point in space, whereas in the second, it changes continuously due to evaporation, dilution or dispersion, or from a combination of the three. The measured change in concentration of tritiated water in the pool system with time due to the dilution with distilled water can be described by the equation

$$\frac{dW}{dt} = -\lambda_w W(t), \tag{2}$$

and the concentration of tritiated water at any time is given by

$$W(t) = W_1 e^{-\lambda_w t}, \tag{3}$$

where W(t) is the concentration of tritiated water at time $\underline{t}$, W_1 is the initial concentration of tritiated water, and λ_w is the evaporation rate constant of tritiated water in the pool system. Substituting W(t) from equation (3) into equation (1), we have the change in tissue-bound tritium concentration in the tissue with time in the pool system,

$$\frac{d(T)}{dt} = \lambda W_1 e^{-\lambda_w t} - \lambda' T(t). \tag{4}$$

Integrating equation (4) and setting T(0) = 0, we have

$$T(t) = \frac{\lambda' W_i}{\lambda' - \lambda_w}\left[e^{-\lambda_w t} - e^{-\lambda' t}\right]. \tag{5}$$

Since tritium bound in tissues is considered to be in a minimum of two compartments, let us assume first that a fraction $\underline{r}$ of the tissue-bound tritium will be rapidly exchangeable and that a fraction $\underline{s}$ will be slowly exchangeable, and second that r + s = 1. The expression becomes

$$T(t) = r\,\frac{\lambda_r W_i}{\lambda'_r - \lambda_w}\left[e^{-\lambda_w t} - e^{-\lambda'_r t}\right] + s\,\frac{\lambda_s W_i}{\lambda'_s - \lambda_w}\left[e^{-\lambda_w t} - e^{-\lambda'_s t}\right], \tag{6}$$

where λ_r and λ'_r are the accumulation and loss constants, respectively, of the tritium in the rapidly exchangeable pool in a clam tissue, and λ_s and λ'_s are the corresponding constants in the more slowly exchangeable pool. Data from experiments in progress show that λ_r and λ'_r are about the same in the clam and are large compared to λ_w. Setting $\lambda_r = \lambda'_r$ and assuming λ_r and $\lambda_r' >> \lambda_w$, the equation simplifies to

$$T(t) = W_i\left[\left(r + \frac{s\lambda_s}{\lambda'_s - \lambda_w}\right) e^{-\lambda_w t} - \left(\frac{s\lambda_s}{\lambda'_s - \lambda_v}\right) e^{-\lambda'_s t}\right]. \tag{7}$$

If we assume that the accumulation constant is equal to the loss constant, then

$$T(t) = W_i \left[\left(r + \frac{s\lambda_s}{\lambda_s - \lambda_w} \right) e^{-\lambda_w t} - \left(\frac{s\lambda_s}{\lambda_s - \lambda_w} \right) e^{-\lambda_s t} \right], \quad (8)$$

where λ_s is the biological turnover rate constant for tritium (identical to that for hydrogen) in the slowly exchangeable pool.

Equation (8) was solved for tissue-bound tritium at varying times in clam tissue after substitution of specific values for s and λ_s. Examples of the changes in shape of the curve from variations in s and λ_s are shown in Figure 8. Increases in $\underline{s}$ result in lower early-time concentrations of tissue-bound tritium whereas increases in λ_s result in higher late-time concentrations. For the viscera, a good fit between the experimental points and the calculated values was obtained with $s = 0.94$ and $\lambda_s = 0.00116$ ($T_b = 600$ days).

Similar calculations were made for the other clam tissues; the calculated curves considered to best fit the data are presented in Figs. 2 and 3. The s and λ_s values used to obtain these curves and the T_b obtained with these λ_s values are presented in Table 2.

Figure 8

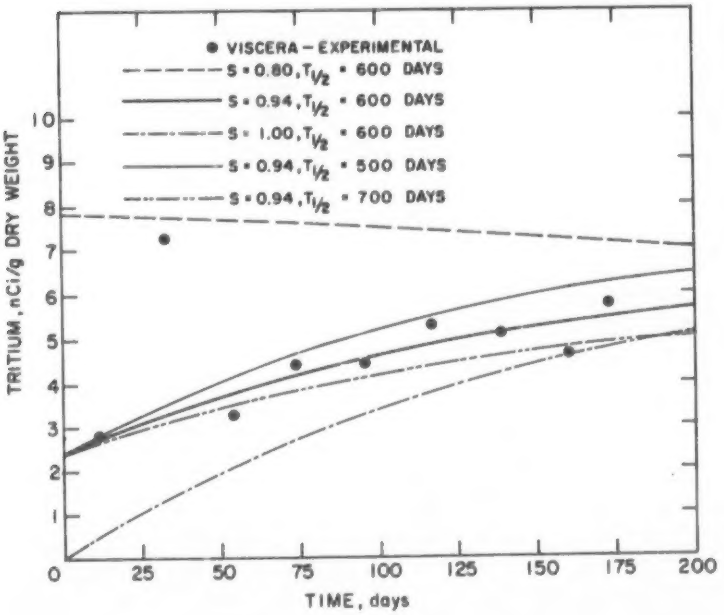


Table 2

Slowly Exchangeable Fractions (s), Turnover Constants (λ_s) and Biological Half-Times for Tritium in Clam Tissues

Tissue	Accumulation			Loss	
	s	$\lambda_s \times 10^4$ (per day)	T_b (days)	$\lambda_s \times 10^3$ (per day)	T_b (days)
Muscle	0.94	5.0	1400	2.4	290
Viscera	0.93	11.0	600	5.8	120
"Peel"	0.92	5.8	1200	3.6	190
Gills	0.86	12.6	550	4.8	150

The values obtained for s were much higher than would have been predicted from data available on mammals. Siri and Evers [7] consider that approximately 30% of the total hydrogen in lean tissue solids is exchangeable. To verify the high s values, exchangeable tritium was determined in selected early-time muscle samples by rehydrating them with nontritiated water. The high s values obtained in the experiment were confirmed. Analyses of samples in various stages of preparation have shown that the exchangeable tissue-bound tritium is very labile; its concentration will change if the samples are heated in an oven or if they are not stored in dry air. Values comparable to those of Siri and Evers [7] were obtained from samples that remained continuously at room temperature in a desiccator. It appears that one fraction of the exchangeable tritium is easily lost from tissues that are somewhat deliquescent due to their high salt content.

The T_b values obtained from the mathematical treatment of the accumulation data were larger than those obtained from the loss data. In adult rats receiving tritium for four months from drinking water only, Thompson and Ballou [8] found the final specific activity of the tissue-bound tritium to be 28% of that of the tissue-free-water tritium in the liver and 20% in the small intestine. Patzer [9] reported, for rats receiving tritium from ingested water only, that the steady-state nonexchangeable tissue-bound tritium was from 17 to 30% that of the tissue-free-water tritium. When the data are expressed in terms of specific activity (pCi/g hydrogen), the specific activities of the muscle and liver were about 30% that of the tissue-free-water tritium. Hatch and Mazrimas [10], in comparable experiments with mice and kangaroo rats, found the specific activity of the tissue-bound tritium in liver to be about one-quarter to one-third that from food and water. In our experiments, the accumulation constants for clam muscle and viscera were about 20% of that of the loss constants. This may indicate that during the accumulation period the animals were obtaining tritium primarily from the water and/or that the conditions during the accumulation period were less favorable than the field conditions for the turnover of hydrogen.

Prediction with the Model

Data on turnover constants and slowly exchangeable fraction can be used in a mathematical model to predict the changes in tissue-bound tritium concentration in clam tissues with time under various environmental conditions. Let us assume a constant rate of release of tritium, so that at a point in space the concentration in water remains the same. In a tissue such as muscle that has a slow turnover of hydrogen, the concentrations of exchangeable and slowly exchangeable tissue-bound tritium will approach that of the water more slowly than do those of viscera with its faster turnover.

Changes in tissue-bound tritium concentration with time after a single release can also be predicted. For a given tissue, the maximum tissue-bound tritium concentration reached will depend on the turnover of water in the ecosystem. In ecosystems where the half-time of the water is short, the concentrations decrease rapidly; in ecosystems where it is long, the concentrations increase gradually before decreasing.

From the model, changes in the ratio of the specific activity in the tissue-bound tritium of the clam tissue to that of the water also can be calculated. After a single tritium release into the ecosystem, the changes in the ratio with time are dependent on the turnover constant of hydrogen in the tissue and the turnover constant of water in the ecosystem. Ratios much greater than unity are obtained; the time after the release at which the ratio exceeds 1 is dependent on λ_s and λ_w. It is apparent that interpretation of field data obtained from ecosystems where the levels of tritium have not been constant must take into consideration the kinetics of the system. Ratios greater than unity may not indicate concentration of tritium but only differences in turnover times in different compartments.

In the calculations performed, it was assumed that the concentration factor for tritium was 1. Higher concentrations of tissue-bound tritium in the tissues are reached if the concentration factor exceeds 1. However, increasing the concentration factor does not result in a linear increase in tissue-bound tritium concentrations, i.e., a concentration factor of 2 does not result in tissue-bound tritium concentrations twice that for a concentration factor of 1.

ACKNOWLEDGMENTS

The authors thank Dorothy Quinn, Jane Ott, and Robert Wikkerink for their technical assistance in the performance of the research. The authors also thank Ann Burton for assistance in the mathematical treatment of the data, Richard Stuart for assistance in the computer program, and the staff at the University of California Bodega Marine Laboratory for their assistance in the maintenance of the experimental animals.

Reference to a company of product name does not imply approval or recommendation of the product by the University of California or the U. S. Atomic Energy Commission to the exclusion of others that may be suitable. This work was performed under the auspices of The U. S. Atomic Energy Commission.

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BEHAVIOR OF TRITIUM IN FISH FOLLOWING CHRONIC EXPOSURE

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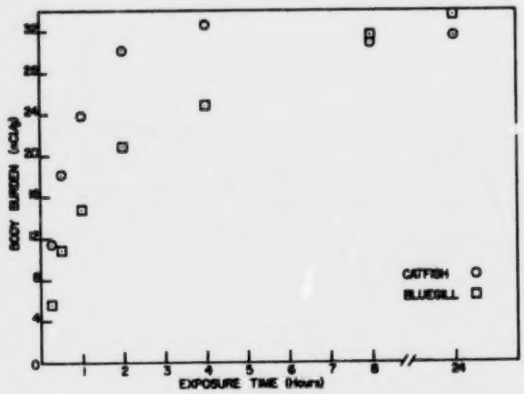
The release of tritium from power reactors presents a need for evaluating the fate of this radionuclide in fish. This information is desirable, not only from the standpoint of game fishing, but particularly with regard to commercial fish farming. The growing of fish, such as channel catfish (Ictalurus punctatus), to edible sizes in cage culture systems is often done in natural bodies of water, and heated effluents from some power plants may be discharged into the waters of commercial fish farms.

The information presented in this report is the result of a preliminary evaluation of the uptake and elimination of tritium by fish grown in tritiated water. These fish were exposed to the tritiated water in 100-gallon tanks which were covered with plastic sheeting to minimize escape of tritium by evaporation. Particulate waste was filtered using pyrex glass wool and dissolved waste products were removed with a bed of charcoal. After passing through the filter, the water was allowed to free fall for approximately 30 cm which provided sufficient aeration to maintain saturated oxygen conditions at all times. The tanks were equipped with a thermostatically controlled heater and a variable flow rate cooling coil which could be operated simultaneously to maintain a wide range of predetermined tank temperatures.

The tritium contained in the fish tissue was analyzed using liquid scintillation spectrometry. Each fish was weighed and homogenized with an equivalent weight of water to a uniform consistency. A weighed aliquot of approximately 100 mg was digested with perchloric acid and hydrogen peroxide. Digestion was allowed to proceed for four hours in an oven at 70°C. Fifteen ml of scintillator were then added to the digested material and counted. The scintillator solution consisted of a mixture of toluene and 2-ethoxyethanol in a ratio of 7:3 respectively containing 4 g 2,5-diphenyloxazole.

The initial study concerned the uptake of tritium in bluegills (Leponis macrochirus) and channel catfish. The fish were exposed to a tritiated water concentration of 40 nCi/ml. The temperature of the water was maintained at 23°C at a pH of 7.5 and a methyl orange (M.O.) alkalinity of 85 ppm. Samples of three fish were taken at 15 and 30 minutes and one, two, four, eight and 24 hours after addition of the fish to the tritiated water. Fish sizes ranged from 4 to 20 g for the catfish and 10 to 65 g for the bluegills. The results are presented in Figure 1, the points representing the mean value of the samples taken at each time period. It can be seen that the uptake of tritium in the catfish has essentially maximized after approximately four hours. The maximum uptake of tritium was reached when the tritium content per unit volume in the body water of the fish was essentially equal to the tritium content per unit volume of the external media.

Figure 1

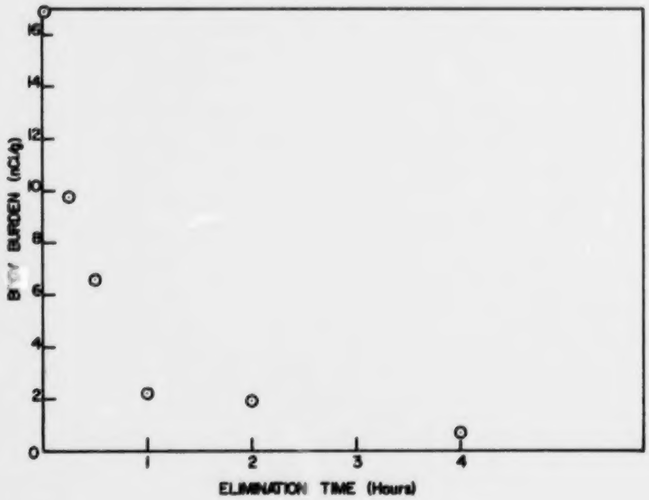


This results in a tritium content per unit mass of tissue equal to approximately 75% of the tritium content of the external media. Since fish tissue is approximately 75% water by weight, this suggests equilibrium was attained between fish body water and the external media. Analysis of fish tissue after removal of the water reveals no organically bound tritium present. Under our experimental conditions it was felt that only after prolonged exposures to tritiated water would significant amounts of organically bound tritium be detected. Approximately 30 minutes are required for the tritium burden in the catfish to reach one-half of its maximum level. This rapid exchange of body water is due to the large flux between the blood and media, although the net increase or decrease of the body fluid is small in comparison.

The time required for the body burden of bluegills to reach one-half of the maximum level was approximately 90 minutes. The difference observed in the uptake rates between the two species is presumably due to the presence of scales on the bluegills. The scales hamper movements of body fluids and, consequently, reduce the surface area available for exchange.

The elimination of tritium from the body of channel catfish was also studied. The fish were exposed to tritiated water at a concentration of 20 nCi/ml for a period of 20 days and then transferred to a tank of uncontaminated water. The water in this tank was maintained at a temperature of 23°C, a pH of 8.3 and an M.O. alkalinity of 250 ppm. The specific conductance of the water was 68 µohm. Samples of three fish were taken at 15 and 30 minutes and at one, two, and four hours after introduction of the fish into the nontritiated water. The channel catfish sizes ranged from 4 to 20 g. The results are shown in Figure 2, each point representing the mean value for each sample. The times required for the channel catfish to eliminate one-half of the body burden was approximately 30 minutes. After four hours, the tritium burden was reduced to minimum detectable levels.

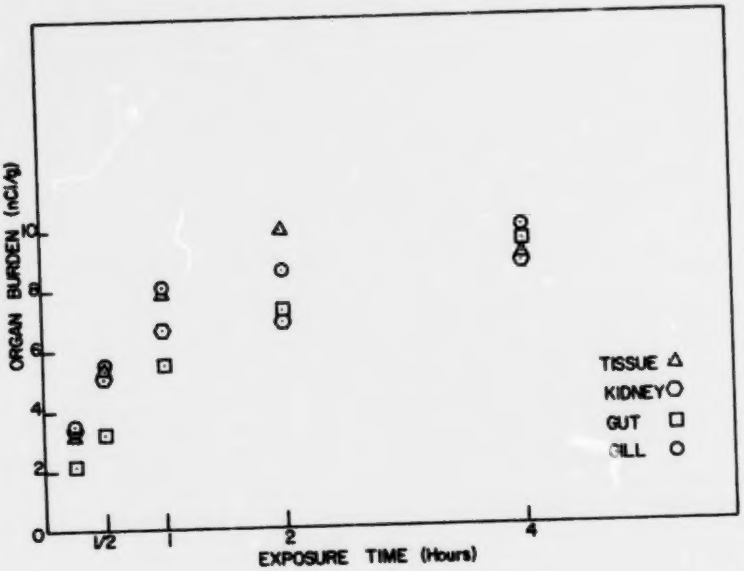
Figure 2



Another experiment was performed to study the uptake of tritium by the gills, kidneys, gut and muscle tissue of channel catfish. This investigation was conducted in water with a tritium concentration of 10 nCi/ml, a pH of 7.7, a water temperature of 26°C, an M.O. alkalinity of 273 ppm, and a specific conductance of 93 μohm. Samples of three fish were taken at 15 and 30 minutes and one, two and four hours. The results are presented in Figure 3. The gills and muscle tissue exhibited the most rapid rate of uptake and the gut had the slowest rate. However, after four hours of exposure, the level of tritium in all tissue was approximately the same. This may be explained by the physiological location of the organs, since the principle flux of water occurs at the gills, followed by distribution throughout the body via the blood.

In summary, it was found that the rate of uptake of tritium was rapid and reached maximum values within approximately four hours, channel catfish exhibiting a more rapid rate than bluegills. The maximum concentration of tritium in both species was approximately 75% of that of the media. Channel catfish exhibited a rapid release of tritium with essentially all the tritium being eliminated in four hours after removal from tritiated water. The rate of uptake of tritium by the gills and muscle tissue was found to be slightly more rapid than for the gut and kidneys.

Figure 3



ACKNOWLEDGMENTS

This investigation was supported, in part, by PHS Training Grant No. T01-EC00064 from the Bureau of Radiological Health, U. S. Public Health Service.

CHAPTER SIX
Certain Environmental Aspects of Tritium

TRITIUM IN THE ATMOSPHERE AND OCEAN

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Tritium was, and is being, produced by natural processes in the atmosphere from the interaction between components of the cosmic ray flux and the nuclei of the atoms of the atmosphere. Before the time of nuclear bomb tests, the total amount of tritium on earth was an equilibrium quantity, in that its global decay rate and global production rate were equal. This equilibrium quantity is now difficult to establish; not enough sampling was performed before the testing of fusion bombs in the atmosphere in the late 1950s and early 60s. However, from a few samples from pre-bomb times, and from estimates of cosmic ray flux and its interactions, it can be estimated that this equilibrium quantity must have been a few kg, possibly up to 8 kg, of tritium. Due to its short half-life, 12 years, only the atmosphere and surface waters may have had measurable tritium concentrations. Between 1954 and 1963 hydrogen bomb testing in the atmosphere released approximately 400 kg of tritium, most of which was injected into the stratosphere. Thus, the cosmic ray production of tritium today plays an unimportant roll in the global mass balance of this isotope [1].

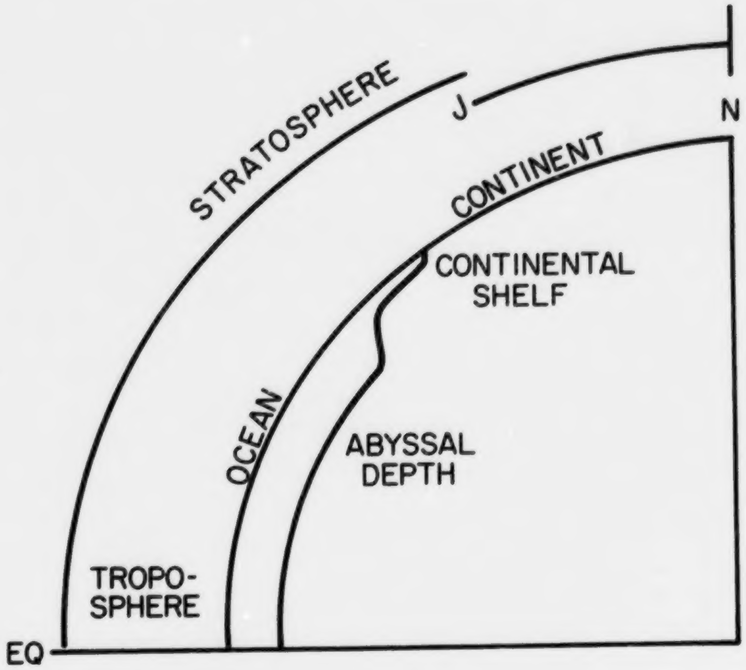
Global Circulation of Water

Most of the surface of the earth is covered by the ocean, and most of the water resides in the deep waters, the so-called "Abyssal Depth," Figure 1. Along the continental coastlines, the depth of the ocean changes suddenly from an average of 4 km in the Abyssal Depth to a few hundred meters on the Continental Shelf.

The lower part of the atmosphere is conveniently divided into two parts: the troposphere, which is lower, and the stratosphere, the upper part. The transition layer between the troposphere and the stratosphere occurs at about 12 km (40,000 feet) and is called the tropopause. This division has come naturally from the fact that the temperature generally decreases, with altitude, to about -56°C at the tropopause. Above that altitude, the temperature does not continue to decrease, but stays constant up to a very high altitude. In the troposphere are clouds, weather, rain, thunderstorms, and rapid mixing. In the stratosphere, disregarding some thin clouds and supersonic transport contrails, the air is clear and, due to the temperature distribution, very stable. The tropopause is lower at high latitudes, and there are one or two discontinuities. In these gaps, the winds blow from west to east; these winds are called the Jet Stream. In the troposphere, water is supplied from the ocean by evaporation.

Figure 1

Water reservoirs of the earth

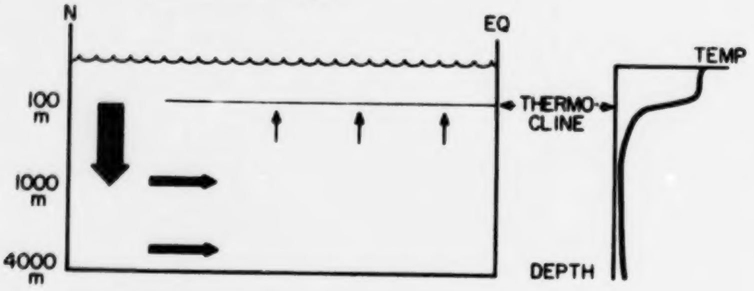


It rains out over land and over the ocean and, if it falls on land, the water ultimately runs back to the sea.

In the ocean, we have an equivalent to the troposphere in that the upper waters are quite well-mixed down to perhaps a few hundred meters, Figure 2. At this depth, called the thermocline, the temperature decreases rapidly with increasing depth. Since warmer water is on top of colder, strong stratification occurs in the thermocline, which thus restrains vertical mixing. The thermocline exists in all warmer waters because of temperature and salinity relationships. In colder water, that is, the Arctic and Antarctic Oceans, the surface water is never warm, and a thermocline does not exist. There the water is sinking and forming the deeper water masses of the oceans. The time scale for the turnover of the deep ocean water is of the order of several hundreds, or even thousands, of years. Therefore, in general, we will expect to find very low tritium concentrations, and no bomb tritium, in the deep ocean.

Figure 2

Generalized picture of ocean circulation in Northern Hemisphere



Pathways of HTO in Nature

In the detonation of the hydrogen bombs, tritium as tritiated water was primarily injected into the stratosphere. From there, slow processes are mixing it down to the troposphere. The processes responsible for this exchange are those that affect the tropopause, most often in the area of the Jet Stream. Another process that sometimes can cause stratospheric water to enter the troposphere is a thunderstorm. Also, possibly very important to mixing, is an occasion when the tropopause is pressed downwards in a trough so sharply that it actually folds over and gets trapped in the troposphere [2]. These mixing processes occur so extensively that the time scale for the transfer of tritiated water from the stratosphere to the troposphere is of the order of a few years. Whenever the water enters the troposphere, it does not remain there. Within a week or so, it rains out and is substituted by evaporation [3].

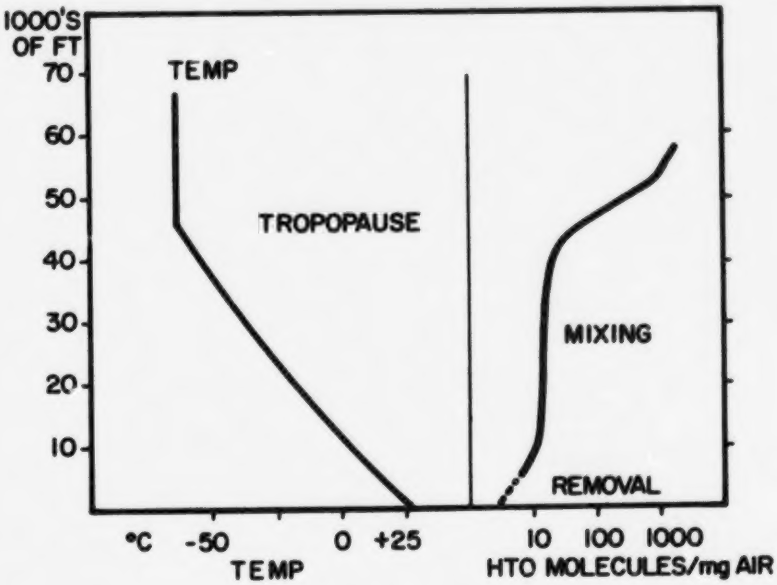
As the ocean is such a large volume of water greatly diluting the tritiated water, it is for all practical purposes the sink for tritium, and the vertical distribution of tritium in the atmosphere should ideally be a steady positive gradient. Figure 3 illustrates the generalized and ideal vertical distribution of temperature and tritium in the atmosphere. Temperature decreases with altitude to the tropopause, as mentioned previously, and the tritiated water content is constant in the same altitude range, due to the rapid mixing. This picture may be true in theory, but it never is in the real world [4,5]. Great variations arise because the air column over any place at any given time consists of layers of different parcels of air, each having a different history as far as exchange with the stratosphere and rain-out processes go.

Actual tritium measurements at altitudes around the tropopause [6,7] have revealed a surprisingly strong gradient (note the logarithmic scale of the tritiated water content in Figure 3). The shape of this gradient can be used for a quantitative attack on the exchange processes across the tropopause.

A process that is very often underestimated is the molecular exchange between liquid and vapor at any water surface; therefore,

Figure 3

Generalized vertical distribution of temperature and tritiated water in the atmosphere



the exchange process removing tritiated water vapor from the atmosphere to the ocean is easily overlooked. The rate of molecular exchange at the water surface can be evaluated by considering the evaporation as the difference between the number of molecules entering and the number being released from the water surface at the same time. At 75% relative humidity, 75 molecules are entering the water surface while 100 molecules are leaving that surface. If the liquid water contains tritium, but the atmospheric vapor does not, the relative loss of tritium from this liquid is three times the relative net evaporation rate of water. If this process is transferred to the ocean, which has a much lower content than the atmosphere above, it appears that molecular exchange is a very efficient scavenging process for pulling the tritium from the surface layer of the atmosphere to the ocean [8-10].

The importance of molecular exchange was directly illustrated in an experiment on Lake Tahoe [11]. Air entering Lake Tahoe in the evening became stagnant in an inversion. Overnight the tritium concentration of the atmospheric moisture of this air parcel was reduced from 157 TU to 68 TU, while its absolute moisture content increased only from 7.1 g/m^3 to 10.4 g/m^3. A similar experiment with tritiated pond water is reported by Horton et al. [12].

The process of molecular exchange occurs not only on lakes and on the ocean, but also on water surfaces in the laboratory and at the air/water interface of animals and plants. Thus an animal with an elevated tritium content in its body fluids, on exposure to tritium-free air, will lose tritium much faster than expected from water loss rates. Or, conversely, if tritiated water vapor is in the laboratory atmosphere, any tritium-free water that we expose to this atmosphere will quite rapidly acquire tritium and become contaminated.

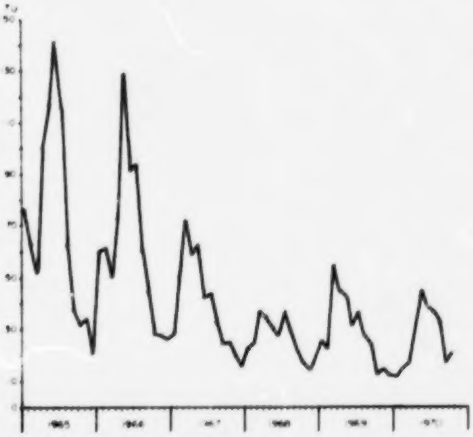
Tritiated Water in Rains

During the past 15 years, a large number of rainwater samples have been analyzed for tritium. Most of them have been collected in a world-wide network of rain-sampling stations, operated by the International Atomic Energy Agency, IAEA, in Vienna, and the World Meteorological Organization, WMO [13,14]. The data have been used as input to studies of hydrology and large-scale atmospheric movements, and also for the clarification of the pathways of tritiated water in nature. Jacobs [1] presents a comprehensive list of references.

There are seasonal variations of the tritium ratio in rains at any given station, similar to the curve in Figure 4, in which the tritium concentrations of monthly rain samples are plotted for Miami, Florida [4,13,14]. We see the characteristic spring or early summer peaks, which have to do with the relocation and effectiveness of the Jet Stream and some other weather phenomena.

Figure 4

Tritium ratio in monthly samples of rain
in Miami, FL, 1965-70

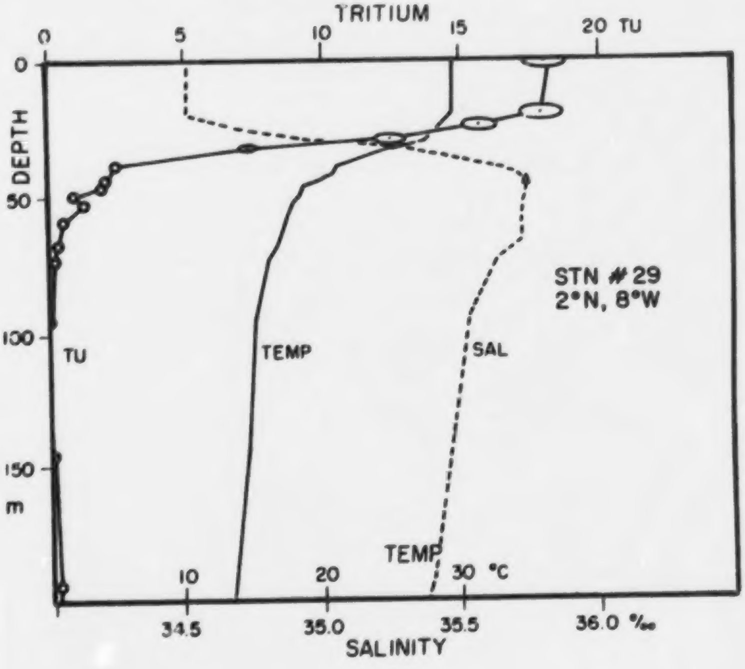


There is an overall removal of tritium from the atmosphere in the summer and fall, to a minimum in the winter. The general decline of yearly averages indicates how the lower stratosphere is being depleted of tritiated water with something like a few years' residence time. This general picture is true for the entire Northern Hemisphere. We find higher average levels at higher latitudes and also higher levels at continental stations than at marine stations. The latter difference depends on the fact that in the marine air the specific activity of the vapor is decreased by molecular exchange with the ocean surface.

Due to the general decrease of tritium levels for the past nine to ten years, it now appears that many continental surface waters have higher tritium specific activities than the atmospheric water vapor at certain times of the year. Therefore, reevaporation of tritium from surface water and plants occurs over the continents, which complicates a study of mixing processes in the troposphere by tritium measurements [5].

Figure 5

Tritium, temperature, and salinity versus depth at a low latitude



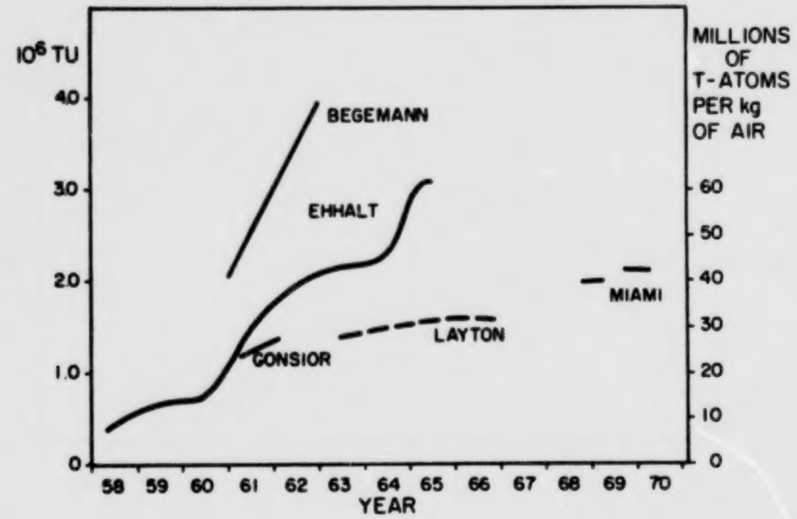
Tritiated Water in the Oceans

The oceans have a thermocline above which mixing is quite fast, and below which time scales are many hundreds of years. This greatly affects the vertical distribution of tritium in the ocean, as exemplified in Figure 5, which is actually a water column very close to the equator in the Atlantic Ocean [15]. This is an unusually shallow thermocline. The temperature and tritium decrease from the surface water downwards.

Measurements of tritium in the ocean can give very good clues to the problems of tracing the oceanic mixing processes and currents [3,16-19]. The water sinking in the Arctic Ocean contains relatively high concentrations of tritium (∿50 TU). It enters the deeper water, and, if a current has a small lateral dimension, i.e., is narrow, we would be able to detect tritium in that current, even if at the bottom of the ocean. We are presently trying to detect tritium in the so-called "Western Boundary Current," which should be a wedge of water flowing along the Abyssal Depth, hugging the wall of the Continental Slope. These measurements require a high degree of sophistication because the specific activities of tritium are extremely low.

Figure 6

Tritium in atmospheric hydrogen. Data smoothed from references 20-23 and results at our laboratory. Conversion from TU to number of T-atoms per kg of air was made assuming 0.5 ppm of hydrogen gas in the atmosphere.

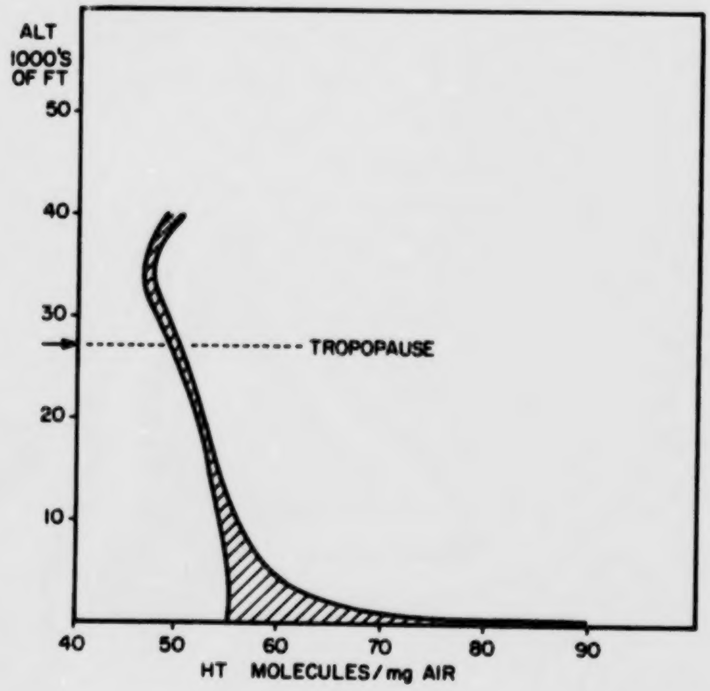


Tritium as an Elemental Hydrogen

Tritium in the form of hydrogen gas in the atmosphere originates from two sources. One is the chemical exchange of tritium between hydrogen and water molecules in the upper atmosphere. The other is the release of tritiated hydrogen gas to the atmosphere at ground level. Data from many authors [20-23] were assembled and plotted in Figure 6, a simplified picture of the tritium gas content of the atmosphere at ground level during the past 15 years. It is striking how this curve, until recently, exhibits a steady increase of elemental tritium in the lower atmosphere in contrast to tritiated water in Figure 4. Samples for this graph were all collected, except the Miami data, at very few locations in heavily industrialized areas.

Figure 7

Vertical distribution of HT. Width of shaded area indicates spread of values.



A new method for sampling atmospheric hydrogen at any place [24] has recently been developed. Thus, tritiated hydrogen in the atmosphere can now be sampled from aircraft and the vertical distribution can be obtained. The first, preliminary results, Figure 7, show a negative gradient with altitude [6]. This indicates the same tendency as the steady yearly increase, namely, that the source of tritiated hydrogen gas in the atmosphere is at ground level. In less academic terms, tritium gas is released, intentionally or unintentionally, by the nuclear industry.

Tritium in Hydrocarbons

Some information is available on the triton content of atmospheric methane and ethane [25-28]. Biological origin for these tritiated molecules is ruled out, since their T/H ratios are much higher than any normal biological substance. Again we look to nuclear industry for a source.

ACKNOWLEDGMENT

This work was supported by grants from the Oceanography and Atmospheric Sciences Sections of the National Science Foundation and also by the U. S. Atomic Energy Commission under Contract No. AT(40-1)-3944.

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THE RADIOLOGICAL HAZARD FROM THE CONVERSION OF TRITIUM TO TRITIATED WATER IN AIR BY METAL CATALYSTS

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The maximum permissible concentrations of tritium gas and tritiated water in air $(MPC)_a$ for occupational exposure are given in ICRP Publication 2 [1] as 2 nCi/ml and 5 pCi/ml respectively. If corrected to take into account the decision to decrease the quality factor for low energy beta emitters from 1.7 to 1.0 [2] these figures become 3.4×10^{-3} μCi/ml and 8.5×10^{-6} μCi/ml. Because of the much greater hazard presented by tritiated water, any release of tritium is usually considered to be in this form, on the assumption that elemental tritium could be converted to tritiated water either by oxidation or by isotopic exchange. However, if the release is of elemental tritium and conversion to tritiated water does not take place, then the hazard is grossly over-emphasized and unnecessary restrictions may be placed on personnel.

Considerable work has already been carried out on the conversion of tritium to the oxide, both by oxidation and exchange processes. Dorfman [3] has studied the reaction between elemental tritium and O_2 at high tritium concentrations and has established that it is of the first order with a reaction rate constant of 1.2×10^{-4} min^{-1} giving a half-time of reaction of about 96 hours. However, at lower concentrations of between 10^{-2} and 1.0 mCi/ml, Casaletto et al. [4] have found a second order reaction rate constant of 6.2×10^{-4} ml mCi^{-1} h^{-1} in dry air. Yang and Gevantman [5] have studied the isotopic exchange between tritium and water vapor in an inert atmosphere and have determined a second order reaction rate constant of 1.5×10^{-3} ml mCi^{-1} h^{-1}. At an initial concentration of 1 μCi/ml the half-time of the oxidation reaction is 6.6×10^4 days and that of the exchange reaction 2.8×10^4 days. Both the oxidation and exchange rates have been found to be independent of either oxygen or water vapor concentration over a considerable range.

From the work referred to above, it is clear that at low concentrations the conversion of elemental tritium to tritiated water in air is a very slow process. However, in a confined space, catalytic reactions may occur on metal surfaces which would increase the conversion rate, and it was to investigate these effects that the following work was undertaken.

Experimental

Glass bulbs of 250 ml capacity were used to contain the tritium gas in all the following experiments. In order to measure reaction rates in the presence of metal catalysts, foils of mild steel, brass, platinum and aluminium with a total surface area of 90 cm^2 were inserted into the bulbs before filling with tritium gas. The foils were chemically cleaned and degreased before use. To investigate the effect of oxide films, foils were cleaned and heated in air at 120°C for one week.

The bulbs were filled with elemental tritium by means of the apparatus shown in Figure 1. The tritium source was supplied by the Radiochemical Centre, Amersham, and consisted of a steel ampoule containing pyrophoric uranium on which was absorbed 100 Ci of elemental tritium. The ampoule was closed by a needle valve and connection made to the vacuum manifold by a glass spiral tube. Tritium was evolved by heating the ampoule to 250°C with a removable wire-wound furnace and reabsorbed by allowing the uranium to cool. The system was pumped out by a two-stage rotary pump via a cold trap; a Pirani gauge was used for pressure measurement.

The bulbs were connected to the vacuum manifold, and the entire system was pumped down to 10^{-3} mm Hg. It was then isolated from the vacuum pump and the pyrophoric uranium heated until the required pressure of tritium was obtained. The glass bulbs were isolated from the manifold and the heater removed from the pyrophoric uranium. Residual tritium was absorbed, and after 30 minutes the needle valve on the pyrophoric uranium ampoule was closed and the manifold flushed out with nitrogen before removing the bulbs.

Figure 1

Apparatus for Filling Reaction Bulbs with Tritium Gas

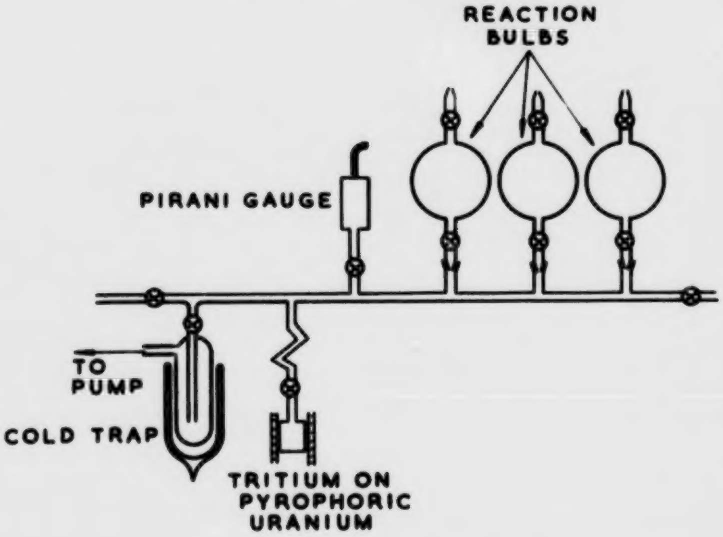
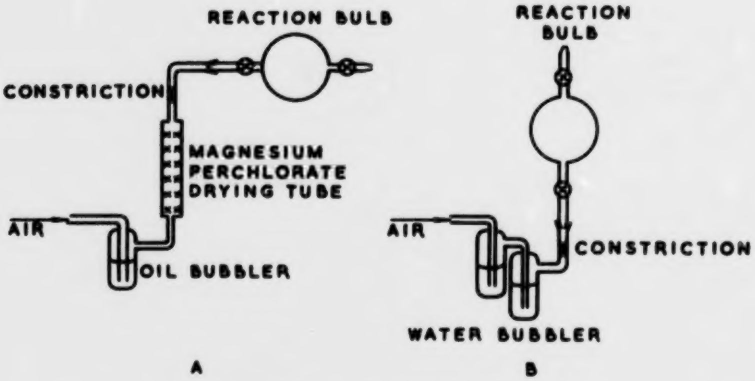


Figure 2

Apparatus for Filling Reaction Bulbs with Dry or Wet Air

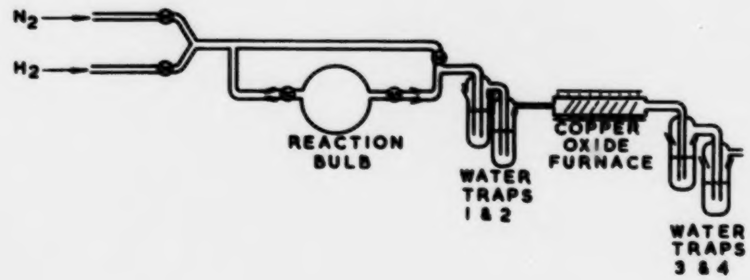


The pressure in the bulbs was brought up to atmospheric with either dry or wet air using the apparatus shown in Figure 2. In Figure 2A, dry air is admitted to the bulb by being drawn through a tube containing magnesium perchlorate. A constriction in the connecting tube between the bulb and drying tube slows down the rate at which air is drawn into the bulb when tap A is opened. A visual indicator of the flow of air into the bulb is given by the oil bubbler. Similarly in Figure 2B, air at 100 percent humidity is admitted to the bulb by being drawn through two water bubblers. In both cases the air flow was controlled so that about 15 minutes were taken to fill the bulb to atmospheric pressure.

The contents of the bulbs were left to react for varying periods of time at room temperature and then analyzed for elemental tritium and tritiated water by means of the apparatus shown in Figure 3. The system was first flushed with nitrogen, with the bulb isolated and the copper oxide furnace heated to 700°C. Nitrogen was then passed through the bulb for one hour, with tritiated water being collected in traps 1 and 2 and elemental tritium (after oxidation to tritiated water in the copper oxide furnace) in traps 3 and 4. A heating tape (not shown) was wrapped round the tube between the furnace and trap 3 to prevent condensation. After an hour, hydrogen was also passed through the bulb to remove adsorbed elemental tritium and the mixed gas flow of nitrogen and hydrogen was maintained for four hours. The copper oxide furnace was allowed to cool, the traps were removed and the concentration of tritiated water in their contents determined by liquid scintillation counting. The glass bulb was washed out with 25 ml of water to remove adsorbed tritiated water and this was also determined by liquid scintillation counting. The sum of these tritiated water measurements represented the total amount of elemental tritium initially present in the bulb and the tritium content of traps 3

Figure 3

Apparatus for Determining Tritium Gas and Tritiated Water in Reaction Bulbs



and 4 represented the amount of elemental tritium remaining after reaction.

Results

In order to check the validity of the method, measurements were first made on the conversion rate of tritium to tritiated water in dry and humid air in the absence of a catalyst. The determinations were carried out at initial tritium concentrations of about 2×10^{-2} mCi ml^{-1}, i.e., within the range of those measurements made by Casaletto *et al*. [4]. These workers found that the initial rate of oxidation of tritium (R_o) was proportional to the square of the initial tritium concentration $(T_2)_o$ and the reaction rate constant $k = R_o/(T_2)_o^2$. This relationship also fits the data for the exchange reaction between tritium and water vapor obtained by Yang and Gevantman [5]. For a measurement of the oxidation rate over a period of time t, Casaletto *et al*. [4] suggest that $k = R/(T_2)_o(T_2)_t$ where one of the tritium terms remains as initial concentration because it represents the radiation intensity which is not changed by the reaction. Hence if the initial concentration of tritium is known, and also its concentration after a known reaction time, then $k = (T_2)_o - (T_2)_t/t(T_2)_o(T_2)_t$

where t = reaction time in hours

$(T_2)_o$ = initial concentration of tritium gas (mCi ml^{-1})

$(T_2)_t$ = final concentration of tritium gas (mCi ml^{-1}) after reaction time t.

Table 1

Oxidation of Tritium in Dry Air

Reaction time (hours)	Rate constant k (ml mCi^{-1} h^{-1})
24	1.4×10^{-3}
96	7.1×10^{-4}
768	3.5×10^{-4}
1464	6.2×10^{-4}
Mean of last 3	5.6×10^{-4}

Table 2

Conversion of Tritium to Tritiated
Water in Air at 100% Humidity

Reaction time (hours)	Rate constant k (ml mCi^{-1} h^{-1})
24	8.1×10^{-3}
72	2.9×10^{-3}
168	2.7×10^{-3}
336	3.1×10^{-3}
Mean of last 3	2.9×10^{-3}

Four determinations of k were made in each case and the results are presented in Tables 1 and 2.

In each series, the first result was omitted in calculating the mean as it is much higher than the subsequent determinations. Small amounts of tritiated water in the elemental tritium used would produce an apparent enhancement in the reaction rate constants over short reaction times and it was considered reasonable to reject these results.

The mean value of 5.6×10^{-4} ml mCi^{-1} h^{-1} for the oxidation of tritium in dry air is in good agreement with the figure of 6.2×10^{-4} ml mCi^{-1} h^{-1} reported by Casaletto et al. [4]. The result in Table 2 of 2.9×10^{-3} ml mCi^{-1} h^{-1} consists of two components, (a) the oxidation rate of tritium to tritiated water and (b) the exchange rate of tritium with water vapor. The latter was found by Yang and Gevantman [5] to be 1.5×10^{-3} ml mCi^{-1} h^{-1} and when added to the oxidation rate constant of 6.2×10^{-4} ml mCi^{-1} h^{-1} [4] gives a combined reaction rate constant of 2.1×10^{-3} ml mCi^{-1} h^{-1}, which is in fair agreement with the value found. The results obtained showed that the experimental technique was satisfactory and confirmed the results obtained by other workers.

Reaction rate constants in the presence of metal catalysts

The rate constants in these experiments were calculated in the same way as those for tritium in air alone, the assumption being that the catalyst would accelerate the reaction but not change its order. This was only confirmed by experiment for steel, in which three separate determinations in dry air for different reaction times gave similar second-order rate constants. It has also been reported [6] that the reaction rate between hydrogen and oxygen on a gold surface is proportional to the square of the hydrogen pressure, and it was assumed therefore that, although the mechanism may be different, the net result of the catalyst is only to increase the reaction rate. The results of the experiments are given in Table 3. The rate constant for steel in dry air represents the mean of three determinations, but the others are single observations.

In dry air there is a marked increase in the reaction rate constant for all metals except brass, with platinum as expected giving the greatest increase. All four metals gave much increased rate constants in the presence of humid air. Again, as expected, the most dramatic increase was with platinum, which increased the rate constant by a factor of 290 over that found in humid air alone.

No significant increase in the rate constant was found for brass in dry air compared with that for dry air alone, in marked contrast to the increase found in humid air. In view of the results obtained for the other metals in dry air, it is possible that the value of 6.0×10^{-4} ml mCi^{-1} h^{-1} for brass in dry air is too low.

Two further measurements were carried out using oxidized brass and steel in humid air. The reaction rate constants were determined as 6.5×10^{-2} and 7.4×10^{-2} ml mCi^{-1} h^{-1} respectively and were thus not greatly different to those found for unoxidized metals.

Table 3

Conversion of Elemental Tritium to Tritiated Water in the Presence of Catalysts

Metal	Rate constant k ($ml\ mCi^{-1}\ h^{-1}$)	
	Dry air	Humid air
Brass	6.0×10^{-4}	8.6×10^{-2}
Steel	4.2×10^{-3}	6.1×10^{-2}
Aluminium	3.9×10^{-3}	9.6×10^{-3}
Platinum	2.4×10^{-2}	8.4×10^{-1}

Table 4

Half-times of Reaction for 10 (MPC)a of Tritium

Catalyst present	Reaction half-time (days)
None	4.2×10^5
Brass	1.4×10^4
Steel	2.0×10^4
Aluminium	1.3×10^5
Platinum	1.5×10^3

Discussion and Conclusions

The measurements described in the previous section are perhaps more clearly understood if the half-periods of reaction are calculated for concentrations of tritium likely to be encountered in operational situations. The ratio of metal surface to volume in the metal catalyst experiments was 0.36 : 1, a condition roughly equivalent to that in a laboratory 20 ft square and 10 ft high. If such a laboratory is considered to be completely lined with the metal being studied, the conversion rate of elemental tritium to tritiated water would be somewhat similar. In Table 4 the half-times of reaction have been calculated for an initial concentration of 3.4×10^{-2} μCi/cm^3 (10 MPC$_a$) of elemental tritium in air at 100 percent humidity.

Thus, in a room completely lined with platinum, one of the best catalysts for conversion of elemental tritium to tritiated water, it would take about four years for half the elemental tritium at an initial concentration of 10 (MPC)$_a$ to be converted to tritiated water. Looking at this situation from another aspect, if the initial release of tritium had remained undetected for one week in such a sealed room, the concentration of tritiated water after this time would only be 1.6×10^{-4} μCi/ml or about 20 (MPC)$_a$. If the room was lined with steel, the concentration of tritiated water after one week would be only about 1.5 (MPC)$_a$. The above figures obviously represent extreme cases where the maximum catalytic surface is assumed in an atmosphere at 100 percent humidity, together with a complete lack of ventilation. In normal circumstances, it is unlikely that this degree of conversion to tritiated water would occur. Other surfaces in a laboratory such as fittings and apparatus would have some effect on the conversion rate, but it is difficult to imagine their having an effect comparable to a total surface area of 1600 square feet of platinum.

It can therefore be seen how important it is to determine the chemical form of tritium in air before imposing unnecessary restrictions on the people at risk. A concentration of 3.4 nCi/ml of tritium in air is equivalent to 4000 (MPC)$_a$ of tritiated water, presenting a very serious hazard, but it is only equivalent to 10

$(MPC)_a$ of elemental tritium, a concentration which may give rise to concern but nothing more. The rate of conversion of elemental tritium to tritiated water is extremely slow under normal conditions at these concentrations. It is therefore wrong to assume that elemental tritium released in an incident will, by oxidation and exchange, be present as tritiated water in significant quantities before it can be dispersed by the simple process of ventilation.

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ENVIRONMENTAL TRITIUM

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The Environmental Protection Agency (EPA) and its predecessor organizations have long recognized the importance of tritium in the environment. Starting from a modest program in the early sixties, the research and monitoring efforts in this field have become extensive. Tritium, being an isotope of hydrogen, is intimately involved in all biological processes, and since it is also radioactive, the possibility of hazard to humans is present, which makes it of substantial interest to the EPA.

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Tritium in the environment originates from natural and man-made sources. In nature, tritium is produced by the interaction of cosmic rays and components of the atmosphere, while the man-made sources are nuclear reactions involved in either weapons development or peaceful uses. Tritium is a fission product as well as a product of fusion reactions and can be produced by many neutron activation processes. The natural equilibrium inventory of tritium is approximately 69 MCi [1]. Weapons tests prior to the signing of the Nuclear Test Ban Treaty contributed about 1700 MCi of tritium to the environment [2]. In the absence of additional thermonuclear explosions, this latter contribution, of course, should decrease at the normal decay rate of tritium. The contribution from power reactors, fuel reprocessing plants, and other nuclear reaction sources is gradually increasing with time. For example, it has been predicted that by the year 2000 the installed electrical generating capacity of nuclear reactors may exceed one TW (terawatt = 10^{12} W) [3,4]. Projections of the tritium accumulation from light water reactor technology indicate that this source may exceed the contributions from natural production and the remaining fallout activity prior to the year 2000 [3,4]. Introduction of fast-breeder nuclear power plants and commercial thermonuclear generating plants may be expected to increase tritium production beyond these estimates.

In view of this predicted increase in the environmental tritium concentration, an adequate and efficient surveillance program is desirable. The analytical techniques used in such a program must be economical, rapid and sensitive. Thus, with the initiation of environmental tritium surveillance, the development of methodology was given a high priority.

Since the discovery of tritium, efforts have been made to develop methods for its determination at environmental levels. The earlier methods used internal proportional counting, together with electrolytic enrichment, for the determination of tritium concentration in rain and other waters. The best internal proportional counters with low backgrounds and high capacities can count these environmental samples without the necessity of enrichment. However, gas counting has many disadvantages when used in a surveillance program, the principal one being the elaborate sample preparation procedures required. Because of this and other factors, the analytical procedure chosen for development was liquid scintillation counting. At the start of the program, the minimum detectable level for liquid scintillation counting was about 4 nCi/l [5]. Modifications of the instrumentation and development of new scintillation liquids reduced this minimum detectable level to 0.2 nCi/l [6,7].

In certain research and surveillance activities, it is desirable to detect lower concentrations of tritium. Increasing the counting time, in a surveillance program where a large number of samples must be counted, seems undesirable. Therefore, to meet the requirements of increased sensitivity, we have developed a large volume scintillation counter which is capable of holding many times the volume of samples contained in the normal liquid scintillation system [8]. We have also developed a tritium monitor in which anthracene-coated rods contained in a cylinder are viewed by two photomultiplier tubes [9,10]. Gases and liquids can be monitored by allowing them to flow through the cylinder. The response of the instrument is linear and has an efficiency of approximately 20 cpm for each nCi/l of tritium in air passing through it.

As mentioned previously, concurrent with the Methodology

Development Program, a Surveillance Program was started [5]. The initial portion of this Program was a study conducted in 1963 to determine the levels of tritium in raw and treated waters collected by the Water Pollution Surveillance Program and by the Institutional Total Diet Sampling Program. Based on the findings of this preliminary study and the reports from other studies, the Tritium Surveillance Program was established in May 1964 to monitor tritium concentrations in the major river systems of the contiguous United States. Adequate coverage was obtained by selecting ten stations from the existing Water Pollution Surveillance sampling stations. Eight of these stations were located downstream from nuclear facilities and two of them served to establish baseline levels [11]. In 1970, this network was replaced by a tritium surveillance system consisting of 70 quarterly drinking water samples collected at the Radiation Alert Network (RAN) Stations and 39 surface water stations [12]. In addition to these networks, EPA operates a precipitation network [13]. Precipitation samples are collected daily and analyzed monthly from eight of the RAN Stations. Precipitation represents a particularly sensitive medium for monitoring tritium in the environment. Any significant addition of tritium into the biosphere can be easily and rapidly detected by analysis of precipitation.

Many other special surveillance activities have been carried out concurrently with these routine national networks. For many years the operation of AEC's Savannah River Plant has been evaluated by comparing the tritium concentrations upstream and downstream from the plant [8].

In a two-year study, surface and ground water samples in the Western United States were analyzed for tritium [14]. Similarly, as part of its Nevada Test Site off-site monitoring responsibilities, the EPA monitors ground waters, milk, atmospheric moisture, and other media for tritium around the Nevada Test Site.

In addition to the water sampling program, food samples collected by the Institutional Total Diet Sampling Network were also analyzed for tritium [15]. As these samples were collected in children's homes, it was also possible to collect urine samples from the residents and analyze for tritium in order to determine any correlations which might exist between the body burden and food intake [16]. These studies were conducted from 1967 to 1968, and in both types of samples a decrease was noted from 1967 to 1968, which agreed with the decreasing trend observed in surface waters and in rain. The ultimate purpose of all these monitoring activities has been to evaluate the possible hazard, to the people, of environmental tritium. The estimation of radiation dose in essentially all cases is based on the following calculation. Assuming a body weight of 70 kg for the so-called standard man, of which 10% is hydrogen, and further assuming that the specific activity of tritium in both the aqueous and organic fractions is the same, then the mrem/year dose equivalent is related to the specific activity of tritium by the equation DE (mrem/year) = $0.1\ Q\ C$(nCi/l). Using a quality factor of 1.7, the dose equivalent can be calculated very simply by multiplying the nCi/l body burden by 0.17. Thus, it was calculated that the dose equivalent for standard man, e.g., in 1967, was 0.15 mrem, and for 1968 was 0.13 mrem. With a quality factor of 1.0, these values are accordingly reduced to 0.9 and 0.8 mrem, respectively. The data collected by all these surveillance programs are published periodically in *Radiological Health Data and Reports*.

Two other human studies that have been conducted by EPA personnel have been the comparison of drinking water and urine tritium concentrations from an area with somewhat elevated tritium levels, and the effective half-lives of tritium in former dial painters. In the first study [17], dealing with comparison of body water and drinking water, it was found that tritium concentrations in urine of different individuals collected on the same date were quite variable, but generally lower than those of drinking water, averaging about 40% of the drinking water tritium concentration. This difference was explained by water intake from sources other than tap water.

In the latter studies [18], on the luminous dial painters, it was found that tritium excretion followed a three-component curve. The effective half-lives for the three components were approximately 10 days, 30 days, and something greater than one year; for convenience one could say that the three components were one week, one month, and one year. These longer components of T_b may have an important bearing on the dose commitment from tritium exposure. These dial painters were located during a study dealing with the occupational aspects of luminous dial painting [19]. Some of these compounds are activated with tritium, and our studies have shown that both the luminous powder and paints made from this powder release tritium into the atmosphere. During these studies, it was found that skin absorption of tritium can play a significant role in increasing the body burden of tritium and so must be considered along with inhalation as a method of exposure.

Studies have also been conducted in conjunction with certain types of Plowshare tests. The first of these was conducted about six years after the cratering test, Project Sedan, to determine the persistence of tritium in the area around the crater. Soil samples taken near the crater indicated that the maximum tritium concentration in soil was approximately at the depth of maximum rainfall penetration. Crops such as radishes, corn, beans, melons, and onions grown in this area had tritium concentrations which were several times those in similar produce bought on the open market. This study indicated that tritium is available in a limited area over considerable periods of time and does not diffuse as rapidly as had been suggested.

For the natural gas stimulation by nuclear explosives experiments, Projects Gasbuggy and Rulison, in addition to tritium surveillance of the gas [21-23] and of plants and animals before and after the tests, a study on the uptake of tritium by swine resulting from domestic use of the stimulated gas was undertaken [24]. The swine were fed a normal human diet cooked on a stove which used tritiated natural gas, and they lived in a chamber which was heated by use of tritiated natural gas. These studies were conducted to relate the body burden of the users of gas relative to certain concentrations of tritium in natural gas.

Other biological studies in progress within the EPA include: The distribution of tritium in water, milk solids and fat portions of milk from dairy cows given oral doses of tritiated water; the rate of migration of tritium through various types of soil; soil-to-plant transfer of tritium in aqueous and organic fractions of plants; and a pilot ecological study in which a circumscribed area of desert was investigated. The soil types were classified and the number and species of plants and animals were identified; and finally, various samples were collected for determination of tritium in the various portions of this circumscribed community.

Other studies have been done in animals, and two of them are worthy of mention. In one study, using rats, tritium body burdens of between 1 and 100 mCi/l were established and maintained throughout pregnancy [25]. At the highest tritium concentration, the findings included microencephaly, sterility, stunting, litter size reduction, and increased resorption of fetuses. These studies have been expanded. Another study deals with the tissue distribution of tritium in rabbits kept over three generations under a constant tritium concentration in their entire intake, i.e., both water and food. This study is also in progress.

An extremely important part of our activities has dealt with tritium release from power reactors and fuel reprocessing plants [26,27]. These studies have been conducted at several nuclear facilities with the cooperation of the facility operators, the AEC and the respective state health departments. These field studies have been conducted at the Dresden Yankee and Connecticut Yankee Nuclear Power Plants, and at the Nuclear Fuel Services' Spent Fuel Reprocessing Plant. Presently, studies are being conducted at the Oyster Creek and H. B. Robinson nuclear plants. Tritium is one of several radionuclides receiving particular emphasis.

Tritium releases from operating power reactors show considerable variability from plant to plant depending on the plant type and operation. We are actively engaged in the review of current operating experience to determine possible areas for improvement and to evaluate effluent characteristics in terms of the waste treatment system design and plant operations. These effluent data are incorporated into our National Environmental Monitoring Program retrieval system and are published periodically [28].

The potential problems associated with the long-term accumulation of long-lived radionuclides in the environment have also been extensively examined. Our projections and assessments of this problem with respect to tritium have been discussed above [3,4]. These evaluations are now considering the fast-breeder reactor fuel cycle and the possible tritium problems associated with thermonuclear power generation. We believe that it is necessary to look at the total impact of nuclear power sources on a long-term basis in addition to the evaluation of each facility and site. These studies have enabled us to develop improved surveillance techniques to better estimate the composition and magnitude of radioactivity release from nuclear facilities and to evaluate critical pathways for population radiation exposure from effluents.

We have attempted to describe certain activities of the EPA related to environmental tritium. Most of the studies mentioned have been published and the data and results are available in more complete form. Our activities related to the environmental problems associated with the development and large-scale peaceful application of nuclear energy are continuing. Through our evaluations, surveillance, and research activities, we hope to insure that this development will have minimal environmental effect.

ACKNOWLEDGMENTS

The assistance of J. A. Lieberman, H. T. Peterson and S. C. Black in the preparation of this manuscript is greatly appreciated.

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THE DOSE TO MAN VIA FOOD-CHAIN TRANSFER RESULTING FROM EXPOSURE TO TRITIATED WATER VAPOR

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Efforts to predict the radiation dose to man as a result of food-chain exposure to tritiated water vapor have been severely handicapped by lack of knowledge concerning the behavior of tritium released to ecosystems, and by the lack of predictive models appropriate for tritium. Attempts to adapt models developed for ^{131}I and radionuclides contained in particulates have been unsuccessful [1].

Predictive failures were evident for Project Rulison, the second experiment in the stimulation of natural gas by nuclear explosives. Results of the Environmental Protection Agency's (EPA) environmental surveillance program [2] and the Lawrence Livermore Laboratory's (LLL) radioecology studies clearly indicate that preflaring estimates of dose resulting from exposure to tritiated water vapor were excessively conservative. Doses were overpredicted by as much as four orders of magnitude [1] due to a combination of conservative assumptions concerning the source term and meteorological and ecological transport.

The purpose of this paper is to provide a more accurate, but also a simple model to predict doses to man resulting from food-chain contamination due to exposure to tritiated water vapor. It incorporates knowledge concerning the environmental behavior of tritiated water which was obtained during radioecology studies at Project Rulison [1] and controlled exposures at Livermore which have been discussed by Koranda and Martin [3]. The model is specifically directed to the prediction of dose from the release of tritiated water vapor from a point source, but most of the model is applicable to any tritiated water vapor exposure.

Dose commitments via inhalation plus absorption and via the ingestion of vegetables, milk, and meat are considered. The model may be entered with a variety of input data; average values of the parameters needed to evaluate the model are provided, but other values more appropriate to a particular situation may be readily substituted.

Washout and rainout are not considered by the model. Theoretical calculations by Chamberlain and Eggleton [4] indicate that washout of tritiated water vapor is relatively unimportant beyond distances equal to a few effective stack heights. Data collected during Project Rulison confirm this prediction [1]. If significant wet deposition of tritiated water should occur, the model may be used to predict dose commitments if the initial tritiated water concentration and residence time in the soil-plant system are known or assumed. A model developed by Kelly and Booth [5] may also be used to predict doses arising from wet deposition, but its use may substantially overestimate the dose if significant loss of tritiated water by evaporation and transpiration occurs.

The model considers only the movement of tritiated water through the aqueous portion of the environment, as not enough information is available to model tritium incorporation and movement through the nonaqueous parts. However, several assumptions are made in the model in an attempt to compensate for the lack of treatment of the nonaqueous portion.

Model Development

Average Concentration of Tritium in Ground-Level Air

For situations where tritiated water vapor is released from a point source at a rate of Q pCi/sec, the simplest means of predicting the average concentration of tritium in ground-level air $\overline{C}$ is the use of the equation [6,7]:

$$\overline{C} = \overline{K}Q$$

where $\overline{K}$ is the weighted mean dispersion constant with units of sec/m^3, Bryant [7,8] provides curves to evaluate $\overline{K}$ as functions of distance from the source and height of release. These values were calculated from data on the frequency of occurrence of meteorological conditions at eight sites in England and Wales. Three sites in Scotland were observed to have essentially similar frequencies of occurrence [8], and Barry [6] has presented arguments for the broad applicability of such data to any geographical site within the middle latitudes.

For a ground-level release, Bryant's values of $\overline{K}$ are 2×10^{-6} and 1×10^{-7} sec/m^3 at distances of one and five km. Any increase in the height of release or distance from the source will reduce the value of $\overline{K}$, but the above value may be used to make conservative estimates of $\overline{C}$.

The use of $\overline{K}$ is only valid for continuous releases. Methods of calculating ground-level concentration following short (a few minutes) or prolonged (six hours) releases are also given by Bryant [8]. If sufficient meteorological data are available, more detailed calculations may be made using methods and data compiled by Turner [9] or Slade [10].

Average Concentration of Tritium in Water of Vegetation

The average concentration of tritium in atmospheric moisture is $\overline{C}$ divided by the absolute humidity H of air in ml/m^3. An essential feature of this model is that the concentration of tritium in vegetation water C_v cannot be higher than $\overline{C}/H$ as a result of exposure to an air activity of $\overline{C}$. While this is obvious, it should be pointed out that models used in the past have predicted values of C_v far in excess of $\overline{C}/H$.

Experimental results indicate that the concentration of tritium in plant water following exposure to tritiated water vapor is actually somewhat lower than C/H. Koranda and Martin [3] exposed eight species of plants to tritiated water vapor for periods of one-half or one hour and found that the ratio of tritium concentration in plant water to that in atmospheric water varied from 0.17 to 0.49. The average value was 0.31; no increase in the ratio was observed when the same species was exposed for one hour as opposed to one-half hour. Studies conducted in conjunction with Project Rulison, where tritiated water vapor was released to a natural environment, provided similar results. Twenty-five exposures of grass were made over time periods of 23 to 51 hours. The average ratio observed was 0.38 [11]. Several experiments of a reverse nature, i.e., where tritiated water was supplied to the nutrient solution, have been reported. The movement of tritium between leaf water and atmospheric water is readily demonstrated as well, where the concentration of tritium in leaf water never equals that in the nutrient medium, but attains an equilibrium ratio of 0.45 to 0.65 [12,13].

The uptake by leaves of tritium contained in atmospheric water is a rapid process. The results of Koranda and Martin [3] indicate that plateau levels are reached by one-half hour. Mason, Hop and Miller [14] indicate that plateau levels are attained within a few minutes.

For predictive model, it is therefore assumed that

$$C_v^o = \frac{0.5\ \bar{C}}{H}$$

during exposure, and that this level is achieved instantaneously. The notation C_v^o is used to indicate the value of C_v during exposure.

After exposure ceases, C_v has been observed to decrease with time in a rather complex manner which appears to depend upon factors such as metabolic activity, length of exposure period, and the extent of soil water contamination.

Several controlled exposure experiments have been conducted at Livermore to investigate the loss process. Exposure is achieved using a portable plastic chamber. Air is circulated in the chamber through a bubbler containing tritiated water [15]. After exposure ceases, the chamber is removed and the plant species are sampled over several days. Samples are immediately frozen on dry ice and subsequently lyophilized. Results observed following a one-half hour exposure of two plant species are shown in Figure 1. Both species

Figure 1

Tritiated water loss curve for two species following a controlled vapor exposure of 0.5 hour. Results are very similar except the concentration of tritium in the second compartment is nearly an order of magnitude different for the two species.

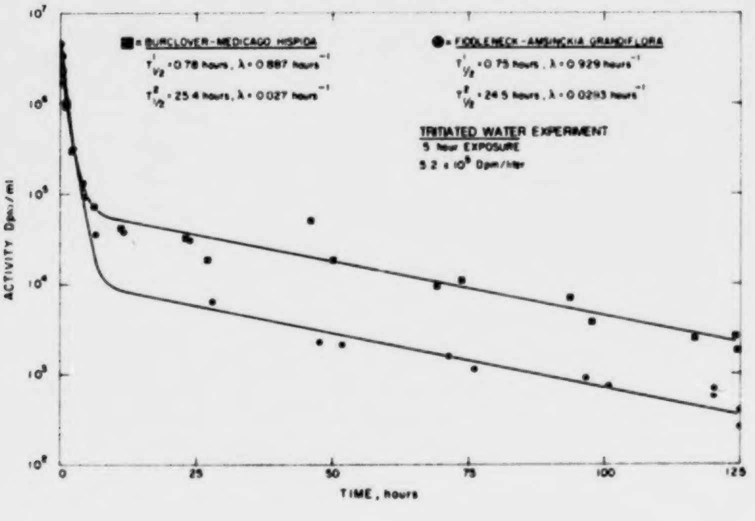

exhibit similar behavior showing two component loss curves, but the concentration of tritium in the second compartment differs by nearly an order of magnitude between the two species. The results of a one-hour exposure of burclover to tritiated water vapor are given in Figures 14 and 15 of Koranda and Martin [3]. Here, three components are clearly evident with the concentration of tritium in the second compartment 4.5 times larger than it was following a one-half hour exposure. Organically bound tritium is eliminated at the same rate as is the third compartment of tritiated water, although it is not clear whether the third tritiated water compartment is derived from organically bound tritium or tritiated soil water [3].

Table 1 summarizes data on the loss of tritium from vegetation

Table 1

Loss parameters observed for the decrease of tritium concentration in vegetation water. The parameters were calculated assuming the loss curves may be represented by a sum of exponentials:

$$\frac{A}{A_0} = \frac{A_1}{A_0} \exp\,(-0.693\ t/T^1_{1/2}) + \frac{A_2}{A_0} \exp\,(-0.693\ t/T^2_{1/2}) + \cdots + \frac{A_n}{A_0} \exp\,(-0.693\ t/T^n_{1/2}).$$

The integral half-time is given by

$$T^{int}_{1/2} = \frac{A_1}{A_0}\,T^1_{1/2} + \frac{A_2}{A_0}\,T^2_{1/2} + \cdots + \frac{A_n}{A_0}\,T^n_{1/2}$$

and represents a single exponential compartment that has an integral equal to the sum of the integrals of each individual component that was measured.

Plant	Exposure Method	Exposure Time (h)	$\frac{A_1}{A_0}$	$T^1_{1/2}$ (h)	$\frac{A_2}{A_0}$	$T^2_{1/2}$ (h)	$\frac{A_3}{A_0}$	$T^3_{1/2}$ (h)	$T^{int}_{1/2}$ (h)
Fiddleneck	Chamber	0.5	0.997	0.7	0.003	24			0.8
Burclover	Chamber	0.5	0.98	0.8	0.02	25			1.3
Burclover	Chamber	1.0	0.908	0.9	0.091	17	0.002	270	2.9
Grass[a]	Project Rulison	45.0	0.25	0.4	0.75	70			53.0

[a] Exposure was late in the year when lower metabolic activity would be expected. Grass plugs were brought into the exposure area 45 hours before the tritium release was stopped. Redeposition of tritiated water from surrounding grass which was more highly contaminated may have significantly influenced these results.

water following exposure to tritiated water vapor. Data from the experiments illustrated in Figure 1, in Figures 14 and 15 of reference 3, and from those conducted in conjunction with Project Rulison [1] are included. These represent the only data known to the authors concerning the persistence of tritiated water in vegetation following exposure to water vapor containing tritium. Ranev and Vaadia [12] took excised leaves of sunflower plants grown in media containing tritiated water and exposed them to an atmosphere saturated with ordinary water vapor. They reported that tritiated water activity was lost with a half-time of about three hours with all activity lost before 24 hours had elapsed.

The data in Table 1 indicate that substantial variation in the integral half-time (defined as the half-time of a single exponential function that has the same integral as the sum of the integrals of the observed components) of THO in vegetation water may be expected due to species variability, length of exposure, and season of the year as it affects metabolic activity. For the predictive model, it is assumed that the integral half-time is one day. This substantially overestimates the measured values for all but the Project Rulison results which may have been atypical due to redeposition from the surrounding environment which contained higher concentrations of tritiated water. However, it appears likely that the concentration of tritium in the second compartment, and therefore the integral half-time, will increase as the length of exposure increases.

The level of C_v after exposure ceases is therefore given by

$$C_v = C_v^o e^{-\lambda_p t'}$$

where λ_p = Disappearance rate of tritium in vegetation water, day^{-1}

= 0.693/integral half-time

$t' = t - T$

t = Time since beginning of exposure, day

T = Time of exposure, day

Concentration of Tritium in Milk Resulting from Forage Contamination

The rate of change of the tritium concentration in milk C_m is given by

$$\frac{dC_m}{dt} = k W C_v - \lambda_m C_m$$

where k = Fraction of the cow's tritiated water body burden per liter of milk, l^{-1} (k is the reciprocal of the volume of the cow's body water pool).

W = Daily volume of vegetation water consumed by a dairy cow, liter/day.

λ_m = Rate of disappearance of tritiated water from the cow, day^{-1}.

During exposure C_v is equal to C_v^o. If exposure is continuous the level of tritiated water in milk at equilibrium, C_m^e, is

$$C_m^e = \frac{k\ W\ C_v^o}{\lambda_m}$$

If the exposure is discrete, C_m during exposure is

$$C_m = \frac{k\ W\ C_v^o}{\lambda_m}(1 - e^{-\lambda_m t})$$

and after exposure ceases at time T, C_m is given by

$$C_m = k\ W\ C_v^o \left[\frac{e^{-\lambda_m t'}}{\lambda_m - \lambda_p} + \left(\frac{1}{\lambda_m} - \frac{e^{-\lambda_m T}}{\lambda_m} - \frac{1}{\lambda_m - \lambda_p} \right) e^{-\lambda_m t'} \right]$$

Concentration of Tritium in Meat Resulting from Forage Contamination

As the concentration of tritium is equal throughout the cow's body water pool [16], the equations used for the concentration of tritium in milk may also be used for meat with appropriate changes in the parameters k, W, and λ_m.

Concentration of Tritium in Milk and Meat Resulting from Bovine Inhalation and Skin Absorption

Bovine inhalation and skin absorption are additional pathways for the introduction of tritiated water into milk and meat. These pathways are not as significant as the ingestion of contaminated fresh forage. However, if cows are fed stored, dry feed, they may be the only significant source of tritium in milk and meat.

During exposure, the activity of tritium in milk or meat is

$$C_m = \frac{2\ k\ B\ C}{\lambda_m}(1 - e^{-\lambda_m t})$$

where 2 is the approximate skin absorption factor for cows (derived from data in references 17 and 18) and B is the bovine breathing rate in m^3/day.

The activity in milk or meat after exposure ceases is given by

$$C_m = \frac{2\ k\ B\ \bar{C}}{\lambda_m}(1 - e^{-\lambda_m T})\ e^{-\lambda_m t'}$$

Concentration of Tritium in Human Body Water

The concentration, C_h, of tritium in human body water from inhalation and submersion during exposure is

$$C_h = \frac{2\ B\ \bar{C}}{\lambda_e M} (1 - e^{-\lambda_e t}),$$

where 2 is the skin absorption factor for both adults and infants, [19,20], λ_e is the rate of elimination of tritium from human body water in day^{-1}, and M is the mass of the body water in grams. If exposure is continuous, the equilibrium value is

$$C_h^e = \frac{2\ B\ \bar{C}}{\lambda_e M} .$$

For acute exposure, the level of C_h after exposure ceases is

$$C_h = \frac{2\ B\ \bar{C}}{\lambda_e M} (1 - e^{-\lambda_e T})\ e^{-\lambda_e t'}$$

The value of C_h from the consumption of vegetables during the period of exposure is given by

$$C_h = \frac{L_v C_v^o}{\lambda_e M} (1 - e^{-\lambda_e t}),$$

where L_v is the daily consumption of vegetables in gram/day. The equilibrium level for continuous exposure is

$$C_h^e = \frac{L_v C_v^o}{\lambda_e M}$$

For acute exposure, the level of C_h after exposure ceases is

$$C_h = \frac{L_v C_v^o}{M} \left[\frac{e^{-\lambda_p t'}}{\lambda_e - \lambda_p} + \left(\frac{1}{\lambda_e} - \frac{e^{-\lambda_e T}}{\lambda_e} - \frac{1}{\lambda_e - \lambda_p} \right) e^{-\lambda_e t'} \right].$$

The concentration in human body water from consuming milk or meat contaminated via forage during exposure is given by

$$C_h = \frac{k\ L_m\ W\ C_v^o}{\lambda_m M} \left[\frac{1}{\lambda_e} + \frac{e^{-\lambda_m t}}{\lambda_m - \lambda_e} - \frac{\lambda_m e^{-\lambda_e t}}{\lambda_e (\lambda_m - \lambda_e)} \right].$$

where L_m is the daily consumption of milk or meat in ml(g)/day. The equilibrium value for continuous exposure is

$$C_h^e = \frac{k\ L_m\ W\ C_v^o}{\lambda_m \lambda_e M} \ ,$$

and for acute exposure, C_h, after exposure ceases is given by

$$C_h = \frac{k\ L_m\ W\ C_v^o}{\lambda_m M}\left[\frac{\lambda_m e^{-\lambda_p t'}}{(\lambda_m - \lambda_p)(\lambda_e - \lambda_p)} + \frac{1}{\lambda_e - \lambda_m}\left(1 - e^{-\lambda_m T} - \frac{\lambda_m}{\lambda_m - \lambda_p}\right)e^{-\lambda_m t'} + \left(\frac{1}{\lambda_e} + \frac{\lambda_m e^{-\lambda_e T}}{\lambda_e(\lambda_m - \lambda_e)} + \frac{\lambda_p(\lambda_e - \lambda_p) - \lambda_m(\lambda_e - \lambda_m)}{(\lambda_m - \lambda_p)\ (\lambda_e - \lambda_m)\ (\lambda_e - \lambda_p)}\right)e^{-\lambda_e t'}\right] .$$

The concentration of tritium in human body water from consuming milk or meat contaminated by two combined pathways, inhalation and skin absorption, is given by the following equation for the time period when exposure is continuing

$$C_h = \frac{2\ k\ B\ \bar{C}\ L_m}{\lambda_m M}\left[\frac{1}{\lambda_e} + \frac{e^{-\lambda_m t}}{\lambda_m - \lambda_e} - \frac{\lambda_m e^{-\lambda_e t}}{\lambda_e(\lambda_m - \lambda_e)}\right]$$

The equilibrium value for continuous exposure is

$$C_h^e = \frac{2\ k\ B\ \bar{C}\ L_m}{\lambda_m \lambda_e M}$$

and for an acute exposure, C_h, after exposure ceases is given by

$$C_h = \frac{2\ k\ B\ \bar{C}\ L_m}{\lambda_m(\lambda_m - \lambda_e)M}\left[(e^{-\lambda_m T} - 1)e^{-\lambda_m t'} + \frac{\lambda_m}{\lambda_e}(1 - e^{-\lambda_e T})e^{-\lambda_e t'}\right] .$$

Dose Rate from Continuous Exposure

If C_h^e for any pathway is in units of pCi/ml (or pCi/g), the dose rate, DR, in rem/year is simply

$$DR = 1.9 \times 10^{-2} \; E \; C_h^e$$

where 1.9×10^{-2} is a dose conversion constant with units of rem-g-dis/(pCi-MeV-Yr). E is the average decay energy in MeV/dis, and the assumption is made that the relative biological effectiveness of the ^3H beta is 1.0 as recently recommended by the International Commission on Radiological Protection [21].

Infinite Dose from Acute Exposure

For acute exposures of duration T days, the infinite dose, D, in rems resulting from the exposure via each pathway is given by the following where A is 5.1×10^{-5} rem-g-dis/(pCi-MeV-day)

$$D = AE \int_o^T C_h \; (t)dt + AE \int_o^\infty C_h \; (t')dt'.$$

The result of this calculation for each pathway is given below

Inhalation: $D = \frac{2AEB\bar{C}}{\lambda_e M} T$

Vegetation: $D = \frac{AEL_v C_v^o}{\lambda_e M} (T + \frac{1}{\lambda_p})$

Milk or Meat
via Forage: $D = \frac{AEkL_m W \; C_v^o}{\lambda_m \lambda_e M} \; (T + \frac{1}{\lambda_p})$

Milk or Meat
via Inhalation: $D = \frac{2kAEB\bar{C}L_m}{\lambda_m \lambda_e M} T$.

The above dose equations include the dose commitment accumulated both during the exposure and after exposure ceases.

Results of the Predictive Model

Typical values of the parameters necessary to use the model are listed in Table 2. Many of the values are well established and were taken directly from published compilations. However, only a few measurements of k and λ_m are available and very few have been reported for beef cattle [16,22-24]. Substantial variations are also to be expected due to environmental conditions [25]. Therefore, to achieve results calculated upon a larger data base, an alternate approach was used. This utilized Comar's compilation of typical data for domestic animals which includes values for body weight, body water, and water intake [26]. The value k is simply the reciprocal of the cow's total body water volume and λ_m is the daily water intake divided by the total body water volume.

Table 2

Numerical values of the parameters necessary to evaluate environmental concentrations and doses resulting from the exposure to tritiated water vapor.

Parameter	Symbol	Value	Ref.
Breathing rate			
Adult	B	20 m³/day	27
Infant	B	5 m³/day	30
Cattle	B	130 m³/day	26
Mass of body water			
Adult	W	43,000 ml	27
Infant	W	6,100 ml	a
Elimination rates			
Vegetation	λ_p	0.7/day	Text
Cattle, dairy	λ_m	0.21/day	26
Cattle, beef	λ_m	0.19/day	26
Adult	λ_e	0.069/day	19
Infant	λ_e	0.22/day	20,28
Reciprocal of cattle body water volume			
Dairy	k	0.0028/l	26
Beef	k	0.0041/l	26
Cattle water intake via fresh forage			
Dairy	W	38 l/day	b
Beef	W	28 l/day	b
Average ^{3}H decay energy	E	0.0063 MeV/dis	29
Human food intake[c]			
Milk			
Adult (20 to 34 a)	L_m	260 g/day	30
Infant (<1 a)	L_m	730 g/day	30
Meat, poultry, fish			
Adult (20 to 34 a)	L_M	270 g/day	30
Infant (<1 a)	L_M	50 g/day	30
Vegetables, fruits, potatoes			
Adult (20 to 34 a)	L_v	600 g/day	30
Infant (<1 a)	L_v	220 g/day	30

[a] Assumes a 10 kg body weight with the same ratio of body water to body weight as that of an adult.

[b] Assumes that dairy and beef cattle need 55 kg and 40 kg, respectively, of green forage per day [26], and that fresh forage is 70% water.

[c] It is assumed for the purpose of dose calculations that each food category is continuously harvested in the exposed area, and is 100% water.

As an illustration of the model predictions, calculations were made for a hypothetical situation where exposure to an air concentration of 10^3 pCi/m^3 of tritiated water vapor is continuous. A value of H of 5 ml/m^3 was chosen to make the calculation. Equilibrium concentrations of tritium in the infant and adult are given in Table 3 as are the predicted yearly doses. The most significant pathways are shown to be inhalation plus absorption, and vegetables for the adult, and milk and vegetables for the infant. Results of a more complex, non-equilibrium calculation are shown in Figures 2 to 4. Calculations were again made with $\bar{C} = 10^3$ pCi/m^3 and $H = 5$ ml/m^3, but the exposure time was taken as 30 days. The predicted levels of tritium in the pertinent environmental media are shown in Figure 2, both during the exposure and 90 days after the exposure ceases. The details of the contributions of each environmental pathway are shown in Figure 3 for the infant and in Figure 4 for the adult.

It is perhaps more interesting to compare the infinite dose predictions for acute exposure to those obtained using other available models. A typical model is that of Ng et al. [31] which, in conjunction with a deposition velocity of 1 cm/sec, was used to predict the potential dose commitments arising from Project Rulison [32]. The results of this comparison are given in Table 4 for a hypothetical one-day exposure to air containing 10^3 pCi/m^3 of tritium.

Table 3

Predicted concentrations of tritium in the body water of man at equilibrium resulting from continuous exposure to air containing 10^3 pCi/m^3 of tritiated water vapor. An absolute humidity of 5 ml/m^3 is assumed.

	^{3}H in Body Water (pCi/ml)		Dose Rate (mrem/a)	
Pathway	Infant	Adult	Infant	Adult
Inhalation, absorption	7.5	13	0.89	1.6
Vegetables	16.0	13	2.0	1.6
Milk, via Forage	26.0	4.4	3.2	0.53
Milk, via Bovine Inhalation	1.8	0.30	0.22	0.036
Meat, via Forage	2.3	5.5	0.27	0.66
Meat, via Bovine Inhalation	0.21	0.51	0.025	0.061
Total	54.0	37.0	6.6	4.5

Figure 2

Predicted levels of tritiated water in pertinent environmental media beginning from the start of a 30-day exposure to air containing 10^3pCi/m^3 of tritiated water vapor. An absolute humidity of 5 ml/m^3 is assumed.

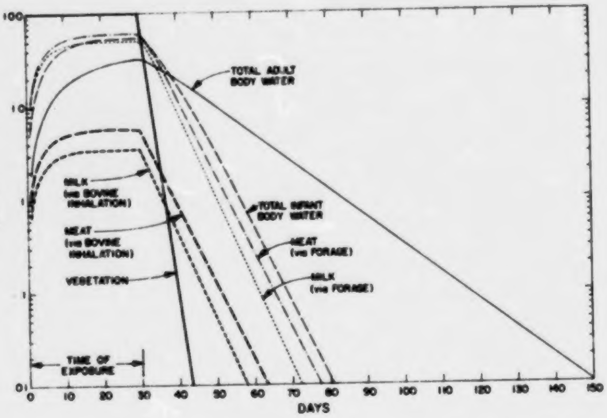

Figure 3

The predicted contribution from each of six pathways to the total tritiated water concentration in the infant's body water. The hypothetical situation is the same as that of Figure 2.

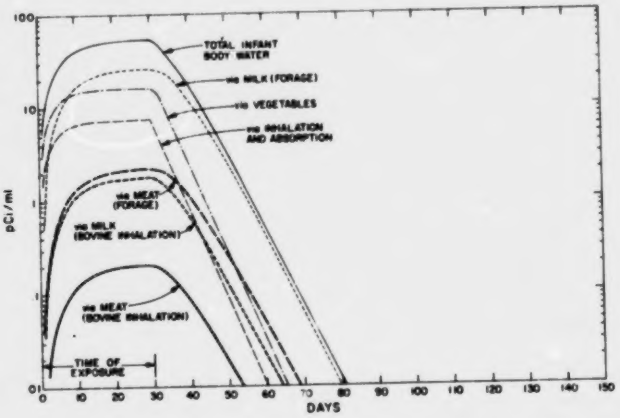

Figure 4

The predicted contribution from each of six pathways to the total tritiated water concentration in the adult's body water. The hypothetical situation is the same as that of Figure 2.

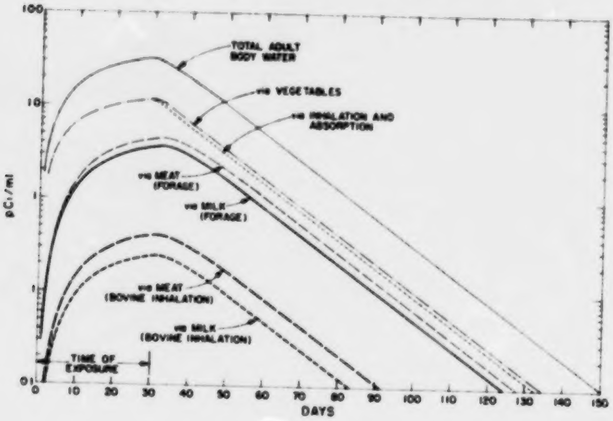

Table 4.

Comparison of predictions for the infinite dose arising from a one-day exposure to air containing 10^3 pCi/m^3 of tritiated water vapor. An absolute humidity of 5 ml/m^3 is assumed for the present model. The typical model used for comparison is that of Ng, et al. [31] with the deposition velocity of tritiated water vapor taken as 1 cm/sec. The model described in this paper predicts the total dose commitment resulting from both the exposure and postexposure periods. The tvpical model considers the dose commitment only in the postexposure period.

Pathway	This Model		Tvpical Model[a]	
	Infant	Adult	Infant	Adult
Inhalation, absorption	2.4 μrem	4.3 μrem	8700 μrem	1300 μrem
Vegetables	13.0	11.0		
Milk, via Forage	21.0	3.5		
Milk, via Bovine Inhalation	0.58	0.098		
Meat, via Forage	1.8	4.3		
Meat, via Bovine Inhalation	0.067	0.16		
Sum	39.0	23.0		

[a] This model was not designed specifically for tritium, but is a general model developed to predict doses arising from the deposition of radionuclides.

Discussion

This model considers tritium moving through an ecosystem only in the form of tritiated water and neglects the organic binding of tritium. However, all food consumed is assumed to be 100% water, and in the dose calculations it is further assumed that tritium is only distributed throughout body water. Under conditions of continuous exposure, the specific activity in the organic portion of foodstuffs would eventually approximate that of water [33,34]; the assumption that foodstuffs are 100% water would then be realistic. The intake of tritium by cattle would be underestimated as only the water of forage is considered. All doses would be somewhat overestimated, however, as all tritium intake is considered to be distributed only in body water rather than the total hydrogen pool. Under acute exposure conditions the assumption that foodstuffs are 100% water overestimates the dose as the specific activity of tritium in the organic compartment will not approach that in water. However, the tritium that is organically bound will leave the ecosystem at a slower rate [3,16,35,36]. These inaccuracies in the model are therefore compensating, but to an uncertain extent. There is not adequate knowledge to construct a model which would include the effects of organic binding throughout a natural, complex ecosystem.

The greatest uncertainty in predicting dose commitments for acute exposures appears to be the evaluation of the parameter λ_p. As already discussed, this parameter appears to vary widely due to biological diversity, metabolic activity, and length of exposure. The value used is probably conservative for very short exposures, but may underestimate the dose for relatively long exposures during periods of low metabolic activity. Kirchmann et al. [35], for example, observed half-times of disappearance of tritiated water in vegetation of one day in summer but five days in winter following the application of tritiated water in a liquid form. However, in this particular experiment only 0.7 to 2.6% of the tritium was still present after 24 hours indicating that the integral half-time is still less than one day. The nature of the decay curve following prolonged vapor exposure during periods of low metabolic activity is, however, an open question. Fortunately, the dose commitment does not vary as a function of $1/\lambda_p$ but as $(T + 1/\lambda_p)$, so as exposures become long and the true value of λ_p becomes questionable, the dose commitments calculated using the model would still be reasonably valid if $T > 1/\lambda_p$.

It appears that the model can be used without modification in rainout situations if the rainout is a few mm or less. Available data indicate that a one-day integral half-time in such situations is still conservative [1,3,35]. If the rainout is sufficient to achieve a significant contamination of subsurface soil water, much longer half-times are to be expected. A discussion of such situations is beyond the scope of this paper. Data pertinent to such situations may be found in references 3, 5 and 37 to 39.

Many of the parameters in Table 2 apply only to hypothetical receptors who are consuming only food which is continually being harvested from the exposed area. In most situations this is an unrealistic assumption, and it is extremely unlikely that a substantial fraction of the predicted dose commitments via the vegetable, meat and milk pathways would ever be delivered following an acute exposure. It has also been assumed that all vegetable water achieves a tritium activity equal to one-half that of atmospheric moisture. Although data are lacking, this is probably true only for leafy vegetables.

Although this model contains several conservative assumptions, the dose commitments predicted by using it are order of magnitude lower than those predicted using other models. This is primarily because other models have used a deposition velocity approach and/or half-times in vegetation water of 14 days [31] or even 85 days [20]. These conservative assumptions were made before adequate data were available indicating the very short half-life of tritium in vegetation. It is now obvious that the use of the deposition velocity approach for the "dry" deposition of tritiated water vapor is inappropriate unless it is considered as a rate constant in competition with a high rate of loss. The greatly lower dose estimates predicted by the use of this model have been verified as conservative on the basis of environmental samples collected by EPA and LLL during the production testing of the Project Rulison well which released tritiated water vapor to the surrounding environment [1].

In all calculations we have neglected the radiological half-life of tritium. This does not introduce a significant error because it is extremely long (12 years) compared to the residence time in the ecological systems considered. This short ecological residence time also indicates that the infinite doses calculated for an acute exposure situation are actually delivered during a relatively short time period, certainly less than a year.

ACKNOWLEDGMENTS

This work was performed under the auspices of the U. S. Atomic Energy Commission.

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GUIDANCE FOR LIMITING ENVIRONMENTAL RELEASES OF TRITIUM

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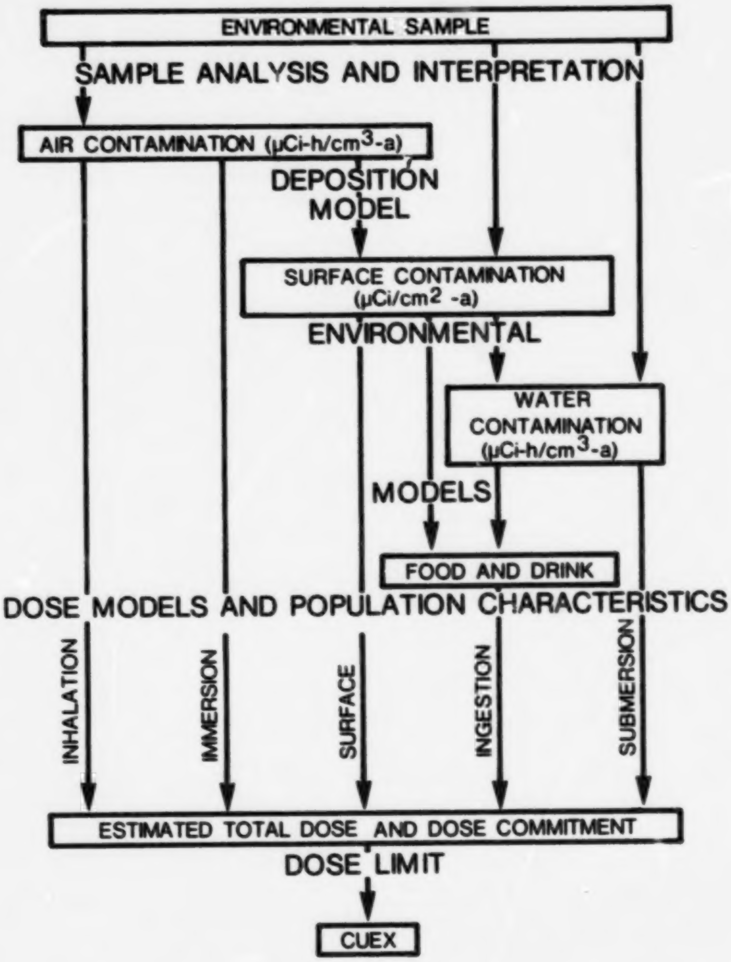
Release of tritium to the environment may result in radiation doses to man. This may occur through a number of exposure modes, each mode having subordinate exposure pathways of potential importance. An exposure "mode" is defined as the manner of exposure, e.g., ingestion, and is differentiated from an exposure "pathway" which is the route of exposure, e.g., pasture-cow-milk-man. Exposure modes of principal importance following an environmental release of tritium are ingestion,

Figure 1

The Cuex Concept


ENVIRONMENTAL SAMPLE
SAMPLE ANALYSIS AND INTERPRETATION
AIR CONTAMINATION (µCi-h/cm3-a)
DEPOSITION MODEL
SURFACE CONTAMINATION (µCi/cm2 -a)
ENVIRONMENTAL
WATER CONTAMINATION (µCi-h/cm3-a)
MODELS
FOOD AND DRINK
DOSE MODELS AND POPULATION CHARACTERISTICS
INHALATION
IMMERSION
SURFACE
INGESTION
SUBMERSION
ESTIMATED TOTAL DOSE AND DOSE COMMITMENT
DOSE LIMIT
CUEX

inhalation, and immersion in contaminated air. To adequately assess the dose commitment from such a release, the potential contribution from each of these exposure modes must be considered. Dose commitment is considered in the assessment of the long-term effects of the release. Dose commitment is the total dose that an individual will accrue within his lifetime as a result of a given radionuclide intake. Throughout the paper, use of the term "dose" should be understood to include "dose commitment" whenever internal exposure modes are involved.

The CUEX Concept

The Cumulative Exposure Index (CUEX) concept is being developed to facilitate realistic assessment of the effect of environmental releases of radionuclides. The goal in developing this concept is to estimate and assess potential dose on the basis of time-integrated radionuclide concentrations measured in environmental sampling media, typically, concentrations in air, water, or on the land surface.

The general structure of the CUEX concept is shown in Figure 1. First, the environmental samples are analyzed and interpreted to estimate the time-integrated radionuclide input to the environment. This input is expressed in $\mu Ci \cdot h/cm^3 \cdot a$ for contaminated air and water and $\mu Ci/cm^2 \cdot a$ for surface contamination. The integration period of one year was chosen to be consistent with the annual dose limit used by present radiation safety standards. Values for the time-integrated radionuclide concentration in air, together with dose models and population data, allow the estimation of dose from inhalation and immersion in contaminated air. Knowledge of the time-integrated radionuclide concentration in air, water, or on the land surface, plus a system of models quantitatively describing the radionuclide behavior between environmental input and man, are required to estimate the potential dose from the other exposure modes. The assessment of an environmental release of radioactivity is completed by comparing the sum of the potential contributions to dose from all five exposure modes with an appropriate annual dose limit.

Calculation of CUEX

Environmental releases of radioactivity usually involve a mixture of radionuclides. Therefore, the radiological safety assessments require estimates of total cumulative doses for a variety of radionuclides and a number of exposure modes. Estimation of Dose Limit Equivalent Concentrations (C) for the individual radionuclides involved is the first step in the CUEX calculation. Calculation of CUEX for a given exposure situation involves C values for each radionuclide present. A C may be calculated for a single exposure mode or any combination of exposure modes. The following formulation is used to calculate a C for exposure via inhalation only, with air as the environmental sampling medium:

$$C_{ijk}^{inh.} = \frac{R_j}{D_{ij}^{inh.}\ 8.33 \times 10^5}\ \mu Ci\cdot h/cm^3\cdot a \qquad (1)$$

where

$C_{ijk}^{inh.}$ = that time-integrated concentration of the i^{th} radionuclide ($\mu Ci\cdot h/cm^3\cdot a$) which, if present in the k^{th} environmental sampling medium under the conditions considered, is estimated to yield a dose for the j^{th} organ, via inhalation exposure only, equal to the selected annual dose limit for that organ,

R_j = selected annual dose limit (rem/a) for the j^{th} organ,

$D_{ij}^{inh.}$ = dose to the j^{th} organ per μCi of the i^{th} radionuclide inhaled (rem/μCi), and

8.33×10^5 = inhalation rate (cm^3/a).

Choice of the inhalation exposure mode as an example is arbitrary, other modes could be selected. The annual dose limit (R_j) should be selected by responsible authorities using recommended radiation safety standards. Each term in Equation (1) may vary as a function of the age of the individual exposed; therefore, a range of C values may be calculated to cover the various age groups in the population. In actual practice, it would be prudent to apply the C appropriate for the "Critical population group" [1]. As the dose and dose commitment contributions for additional modes of exposure are estimated, the C for the initial exposure mode (inhalation in this case) is reduced in the following manner:

$$C_{ijk} = C_{ijk}^{inh.} \frac{R_j}{R_j + \sum_{\ell=1}^{n} D_{ij\ell}} \qquad \mu Ci\cdot h/cm^3\cdot a, \qquad (2)$$

where

C_{ijk} = that time-integrated concentration of the i^{th} radionuclide ($\mu Ci\cdot h/cm^3\cdot a$) which, if present in the k^{th} environmental sampling medium under the conditions considered, is estimated to yield a dose for the j^{th} organ, via all exposure modes, equal to the selected annual dose limit for that organ,

$C_{ink}^{inh.}$ = that time-integrated concentration of the i^{th} radionuclide ($\mu Ci\cdot h/cm^3\cdot a$) which, if present in the k^{th} environmental sampling medium under the conditions considered, is estimated to yield a dose for the j^{th} organ, via inhalation exposure only, equal to the selected annual dose limit for that organ,

R_j = selected annual dose limit (rem/a) for the $j^{\underline{th}}$ organ, and

$D_{ij\ell}$ = dose (rems) to the $j^{\underline{th}}$ organ from the $i^{\underline{th}}$ radionuclide in the $\ell^{\underline{th}}$ exposure mode (ℓ denotes any exposure mode other than inhalation) for an exposure equivalent to $C_{ijk}^{inh.}$.

The Cumulative Exposure Index for a given environmental release of radionuclides is calculated in the following manner once the Cs have been calculated and the time-integrated radionuclide concentrations in the environmental sampling medium have been estimated:

$$\mathrm{CUEX}_j = \sum_{i=1}^{n} \frac{E_{ik}}{C_{ijk}} \tag{3}$$

where

CUEX_j = a numerical guide indicating the relative significance (dose estimate ÷ dose limit) of measured environmental radioactivity on the basis of the estimated total dose in the $j^{\underline{th}}$ organ for all radionuclides and all exposure modes,

E_{ik} = time-integrated exposure (μCi·h/cm^3·a) for the $i^{\underline{th}}$ radionuclide in the $k^{\underline{th}}$ environmental sampling medium, and

C_{ijk} = that time-integrated concentration of the $i^{\underline{th}}$ radionuclide (μCi·h/cm^3·a) which, if present in the $k^{\underline{th}}$ environmental sampling medium under the conditions considered, is estimated to yield a dose for the $j^{\underline{th}}$ organ, via all exposure modes, equal to the selected annual dose limit for that organ.

Selection of the final CUEX may require calculation of several CUEX_j values. The primary reason for multiple calculations is the following: Evaluation of an exposure involving a mixture of radionuclides is complex because dose contributions vary among reference organs and among radionuclides, and the result is a spectrum of reference organs irradiated in varying degrees by each radionuclide. For a conservative assessment of the exposure situation, the largest CUEX_j among the reference organs considered would be selected. Following the ICRP recommendations regarding significant exposure of several organs [1], the exposure of man for a given environmental release of radioactivity would be considered excessive if any one CUEX_j value is greater than 1.0, or if three or more CUEX_j values each are greater than 0.5. The objective in controlling environmental releases of radioactivity on the basis of this concept is to reduce the final CUEX as far below 1.0 as practicable.

Application of CUEX Concept

In practice, it will be necessary to calculate Cs, and subsequently CUEX, for each site. This is because the releases will vary from site to site, and the environmental factors affecting dispersion and bioaccumulation will be unique to each situation, as will the demographic factors which affect the doses or dose commitments to people. Health physicists, or radiological health specialists, faced with a potential population exposure situation requiring assessment, would calculate and apply their own Cs and CUEX with input data and interpretations that specifically apply to the situation in question.

Calculation of a CUEX for Tritium Oxide

The important exposure modes for tritium oxide are ingestion, inhalation, and immersion in contaminated air. The small decay energy excludes the contaminated surface mode, and the short exposure probability and dilution excludes the submersion mode in lakes and streams. For the purpose of these calculations the simplifying assumption was made that tritium oxide distributes uniformly over the body-water pool, resulting in whole-body exposure.

To assess the radiological dose for tritium within the CUEX concept, we shall consider the possible consequences of an excavation experiment at The Nevada Test Site performed in late April. The tritium oxide released to the main cloud reaches the ground in rainfall at 1500 km (Omaha, NE) or 3000 km (Coshocton, OH) using a specialized environmental model for tritium oxide. Both Omaha and Coshocton have been on the cloud trajectories for some previous events at the Nevada Test Site.

The environmental sampling medium is rainfall and the concentration in $\mu Ci/cm^3$ and total rainfall in cm is required. We shall assume only one such test per year so our time-integrated environmental input is most conveniently expressed as $(\mu Ci/cm^3)(cm/a) = \mu Ci/cm^3 \cdot a$. The average concentration of tritium in air at ground level could be measured and the dose from inhalation and immersion calculated during cloud passage. However, this transient exposure is negligible compared to subsequent possible exposures.

A heavy rain is required for maximum deposition, although the model for subsequent exposures is nearly independent of the amount of initial rainfall.

The tritium environmental behavior model [2] considers tritium deposition as rainfall from a cloud containing both HTO and H_2O. The initial distribution in the soil is determined by the void volume in the soil and the amount of rainfall. Further distribution is achieved by interdiffusion, translation by subsequent rainfall, and loss by evapotranspiration.

Usually a material balance of total water in the soil root zone is maintained (rainfall = percolation + evapotranspiration), although water gain or loss conditions can be simulated. The average concentration of tritium in the soil root box is determined over space and time, and then its transfer to man as food is considered.

The space and time-dependent diffusion equation for the movement of tritium in the soil from an initial concentration inserted over a finite depth is solved. Ground surface is taken as the x = 0 plane and the concentration of tritium changes in space only with x, the depth in the ground. The diffusion equation and the appropriate boundary and initial conditions are

$$\frac{\partial c(x,t)}{\partial t} = D\,\frac{\partial^2 c(x,t)}{\partial x^2} - v_2\,\frac{\partial c}{\partial x}\,(x,t) - \Sigma_a\,c(x,t) \tag{4}$$

$$c(\infty,t) = 0,\quad -D\,\frac{\partial c(x,t)}{\partial x}\bigg|_{x=o} + v_2\,c(x,t)\bigg|_{x=o} = 0 \quad \text{and}$$

$$c(x,t) = C_o\,\frac{f}{h} \quad \text{for } 0 < x \leq h \text{ at } t = 0$$

$$c(x,t) = 0 \qquad \text{for } x > h \qquad \text{at } t = 0$$

where $c(x,t)$ is the concentration of tritium in Ci/cm^3, D is the interdiffusion coefficient of water (cm^2/day), h is the depth of the initial tritium pulse (cm), f is the fraction of water in the soil, and C_o is the initial deposition (Ci/cm^2). The value v_2 is the average velocity into the ground (cm/day) from subsequent uncontaminated rainfall (v_2 = rainfall rate/void fraction) and Σ_a is the fraction of total water (HTO + H_2O) removed from soil root box per day (day^{-1}).

The solution to this equation was programmed on a digital computer. The following parameter values were chosen for Omaha on the basis of average rainfall and a material balance for water: $D = 1 \times 10^{-5}$ cm^2/sec, $C_o = 10^8$ pCi/cm^2, h = 0 or 31 cm, v_2 = 0.048 cm/day, f = 0.3 and $\Sigma_a = 0.32 \times 10^{-2}$ day^{-1}. For Coshocton, three of these parameters were adjusted to the following values to be consistent with lysimeter data reported [3] at the site for corn, wheat, and pasture grasses: $C_o = 10^7$ pCi/cm^2, v_2 = 0.42 cm/day, and $\Sigma_a = 0.367 \times 10^{-2}$ day^{-1}. For both locations, the initial soil root box was 230 cm deep, containing 69 cm of water. As the depth of the soil root box was decreased to determine the effect of depth on environmental behavior, Σ_{ad} was calculated as $\Sigma_{a230} \times 230/d$, since evapotranspiration is independent of soil root depth.

After $c(x,t)$ is calculated for a set of discrete values for x and t, the average concentration of tritium in Ci/cm^3 in the volume from X = 0 to x = root depth, $\overline{c}(t)$, is determined for each discrete time.

The transfer of tritium to man by ingestion is based on the assumption that equilibrium exists between the average concentration in the soil water over the root zone and the food crops, milk, and animal tissue consumed by man. A 2000 g/day intake of food is used and 1 μCi of tritium ingested is considered to deliver a whole-body dose of 0.136 mrem to an adult. Inter-age-group variation in the dose resulting from tritium ingestion is expected to be less than a factor of 2 [4]. Calculated C values for tritium oxide deposition using this model and an annual dose limit of 170 mrem are shown in Table 1 for various soil root depths.

Table 1

C for Tritium Ingested as Food

$\left(\frac{\mu Ci}{cm^2 \cdot a}\right)$

(Dose Limit taken as 170 mrem/a)

Root Depth (cm)	Omaha, Nebraska		Coshocton, Ohio	
	Light Rain (h= <1)	10 cm Rain (h=31)	Light Rain	10 cm Rain
9	0.099	a	0.148	a
25	0.109	a	0.184	a
45	0.114	0.126	0.197	0.258
65	0.119	0.126	0.202	0.239
85	0.126	0.131	0.205	0.231
110	0.136	0.141	0.207	0.226
150	0.155	0.159	0.212	0.224
190	0.175	0.179	0.225	0.232
230	0.176	0.200	0.243	0.249
Average	0.137	0.152	0.203	0.237

[a] Do not contain total HTO

The variation of our C (ingestion) values is less than a factor of 2 over a wide range of soil root depth and with rainfall from 0 to 10 cm. This is because the environmental half-life varies inversely with the depth of the soil root zone while percolation and diffusion play secondary roles. We shall arbitrarily chose the average C values for surface deposition in each case.

Climatological data from both sites for May through September show an average of 13 g of water per cubic meter of air at ground level. Assuming this is all supplied by evapotranspiration from the soil root box (i.e., no mixing with upper air) at the tritium concentrations specified by our C (ingestion) calculations, we can make a very conservative approximation of the average concentration of tritium oxide per cubic meter of air. The potential dose is then calculated using reference data [4]. This dose from inhalation and immersion is 22 mrem/a from each of these modes at both locations.

The C (ingestion) values must be reduced as previously discussed to account for these modes, so

$$C_{\text{Omaha}}^{\text{HTO net}} = 0.109 \frac{\mu Ci}{cm^2 \cdot a}$$

$$C_{\text{Coshocton}}^{\text{HTO net}} = 0.161 \frac{\mu Ci}{cm^2 \cdot a}$$

The possible tritium oxide deposited in rainfall at various distances from the main cloud formed by a 1-megaton excavation test may be estimated using the Crawford technique [5]. These are 10^{-2} $\mu Ci/cm^2 \cdot a$ and 10^{-3} $\mu Ci/cm^2 \cdot a$ at Omaha and Coshocton, respectively.

Using these values

$$\mathrm{CUEX}^{\mathrm{HTO}}_{\mathrm{Omaha}} = \frac{10^{-2}}{0.109} = 0.0917$$

$$\mathrm{CUEX}^{\mathrm{HTO}}_{\mathrm{Coshocton}} = \frac{10^{-3}}{0.161} = 0.0062$$

Using the definition of CUEX (dose estimate : dose limit), these values correspond to whole-body dose estimates of 16 mrem/a and 1 mrem/a for residents of Omaha and Coshocton, respectively.

In order to complete the radiological assessment of such an excavation experiment, a similar treatment would be applied to all significant radionuclides deposited and the final CUEX values, Equation 3, would reflect the estimated total dose to man.

ACKNOWLEDGMENTS

Research was sponsored by the U.S. Atomic Energy Commission under a contract with Union Carbide Corporation.

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THE MOVEMENT OF TRITIUM IN ECOLOGICAL SYSTEMS

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The detection of tritium as a contaminant in the biosphere occurred shortly after the advent of the thermonuclear era and the detonation of the first thermonuclear explosive [1-3]. Other sources which may release tritium to the environment are tritium separation plants, nuclear power reactors, nuclear fuel processing plants, and particle accelerators. A future source of tritium is the controlled thermonuclear or fusion reactor whose fuel is tritium and deuterium [4]. It seems inevitable that the biosphere will acquire an increasing burden of tritium as the nuclear age unfolds, and detailed information on the behavior of tritium in many kinds of ecological systems will be needed to determine the hazards associated with operations releasing tritium into an uncontrolled environment.

Jacobs [5] calculates that by the year 2000 in the free-world nuclear power economy, the accumulated fission-product inventory of tritium in the biosphere will be 9.6 X 10^7 curies. However, it will not be until 1990 that the natural rate of tritium production, at 4 to 8 MCi per annum with a steady state inventory of 70 to 140 MCi, will be exceeded.

The capacity of the biosphere for dilution of tritium would seem to be almost infinite. On the other hand, the rapid interaction of tritium with biological and physical systems of an environment may partially counteract this potential dilution if the dynamics of tritium in those systems is such that significant incorporation and retention occurs. When tritium is released into an environment, it moves through the local ecosystem into hydrological compartments until it reaches the oceans. Perhaps potential dilution should not be calculated at the level of the biosphere, but at the ecosystem level where tritium first comes into contact with biological and physical systems in the environment.

In these discussions, the effects of an exposure of the ecological system or its major components to tritium as tritiated water will be described. Releases of elemental tritium may be expected from some facilities, but a fraction of the released elemental tritium becomes oxidized to tritiated water, approximately 0.2 to 3.0% per day [6], and therefore, the situation is essentially a tritiated water release. It must be noted, however, that absorption of elemental tritium does occur to a limited extent in mammals [7] and also in vascular plants [8].

Tritiated water may be released into an ecological system either in the form of liquid or as vapor. Because water changes phases easily and often in ecological systems, the tritium release may also change its phase after release in the environment. For example, after tritiated water is deposited on plants and soil, it may vaporize as evaporation and plant transpiration. A vapor release may be deposited as water if vapor interacts with rain or is cooled [9]. It is necessary to know the behavior of both vapor and liquid releases of tritiated water because either form can constitute the dominant mode of release from an installation. As we shall demonstrate later, the form of the release will determine the persistence or half-time of the tritiated water in that ecological system.

In any terrestrial ecological system, there are three main levels through which water, nutrients, trace elements, and contaminants flow. These are the substratum or soil, the plants growing in that substratum, and the resident herbivores and carnivores. Man is usually the resident herbivore/carnivore considered in this type of discussion. Anspaugh *et al*. discuss human food chain relationships of tritium deposition in ecological systems in this volume. Emphasis is placed on soil and plant tritium interrelationships in this paper because human and mammalian physiology have been discussed elsewhere in this book.

The soil system

The movement of tritium as tritiated water in the soil will be affected by three salient parameters: (1) structural characteristics of the soil, (2) subsequent additions of water to the soil, and (3) soil water present in the soil before tritium application or exposure. These general statements are supported by the research of various investigators which will be discussed in the following text. The bulk density and permeability of the soil will determine the rate of penetration of the tritiated water pulse and the amount of tritium entering surface runoff. High clay soils may be quite impervious to through-flow of water, and a pulse of tritium in this type of soil system would have a different rate than one deposited on a well-drained podzolic soil.

The water volume in the soil before tritium deposition would seem to be a potential dilutant but, because mixing within the soil profile is not complete, the tritium pulse remains relatively discrete as it moves lens-like through the soil, under the influence of subsequent depositions of rainfall or irrigation. In porous, sandy soils receiving a low rainfall rate, such as in the desert, diffusion of soil water vapor may play a significant role in movements of tritiated water. The intensity and distribution of rainfall or irrigation after tritium deposition will determine the half-time of tritium in the soil system, with secondary effects being exerted by the physical characteristics of the soil. These general statements will be demonstrated by experimental data obtained in field studies concerned with tritium movement in ecological systems or their components.

Zimmerman et al. [10] provide experimental data which describe the behavior of a pulse of deuterated water in soil systems. Their data show that an application of labeled water onto the surface of the soil will be eluted into the soil profile as successive additions of water are received. The labeled water pulse moves in sheet-like fashion through the profile, blurred somewhat by diffusion effects but still discernible as a tagged layer. The authors state that the velocity of the tagged water stratum depends upon the average velocity of all the water in the soil; therefore, the more water that is present in the soil, the more slowly the tracer moves. Fine grain soils showed an average draining rate of less than one meter per year.

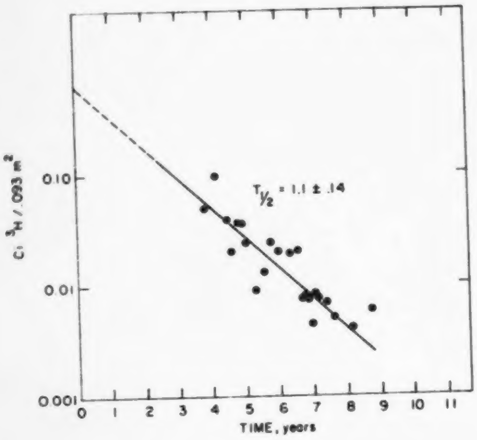
Kline and Jordan [11] studied tritium movement in tropical montane soils in Puerto Rico. In their studies, tritium was applied to the forest floor and then as rainfall eluted the tritium pulse into the profile, soil water samples were collected at the organic matter-solum interface and at a depth of 18 cm in the mineral soil. The litter layer showed a two-component loss of tritium while the mineral soil at 18 cm demonstrated a logarithmic die-away of tritium with a half-time of 16.3 days.

A more detailed study of the movement of a tritium pulse in a tropical rainforest soil was conducted by Jordan et al. [12] at the same site [11] in Puerto Rico. The wave-like movement of the tritium pulse through the soil profile was clearly observed in these studies by simultaneously collecting soil water samples at several depths. Zero-tension lysimeters [13] were installed in the soil profile to intercept soil water percolating downward. The significance of this characteristic movement of soil water and tritium in the soil is that it produces an extended half-time of tritium in plants growing in the soil. As the tritium pulse passes through the soil profile and the rhizosphere of the plant, successively deeper portions of the root system are exposed, and the plant thus "sees" the tritium pulse for a longer period of time than if the tritium entered the plant stem or foliage only. The plant tritium half-time and the soil-plant tritium half-time were separated in these studies by two separate experiments [14]. The half-time of tritium in the tree was determined by sampling leaves after tritiated water had been injected into the trunk. The tree half-time for tritium was 6.6 days while the soil-tree half-time was 43±4 days.

Koranda and Martin [15] and Martin and Koranda [16] have studied a tritiated soil profile at Sedan crater in the Nevada desert for the past five years. Approximately one million curies of tritium were dispersed into five to six million tons of earth moved by the Sedan detonation. These studies have determined that the environmental half-time of tritium in the dry desert environment of the Nevada Test Site, with approximately 75 mm of rainfall per year, is on the order of 1.1±0.2 years. This measurement was obtained by sampling the soil or ejecta to a depth of 1.8 to 2.4 meters, integrating the tritium concentrations in the soil profile, and expressing these data as activity per unit area. The data, expressed as µCi per 0.093 $meter^2$, were then compared in time to obtain the environmental half-time estimation. This half-time measurement is affected by evaporative losses, plant uptake and transpiration, but little if any losses to the ground water occur. Soil materials at Sedan crater are sandy and contain a small amount of clay. Figure 1 shows the environmental half-time data for tritium at Sedan crater during the five years of these measurements. It should be noted that the original conditions at Sedan crater, just after the detonation, probably do not represent a surface deposition of tritium.

Figure 1

Environmental Half-time of Tritium in Sedan Crater Ejecta

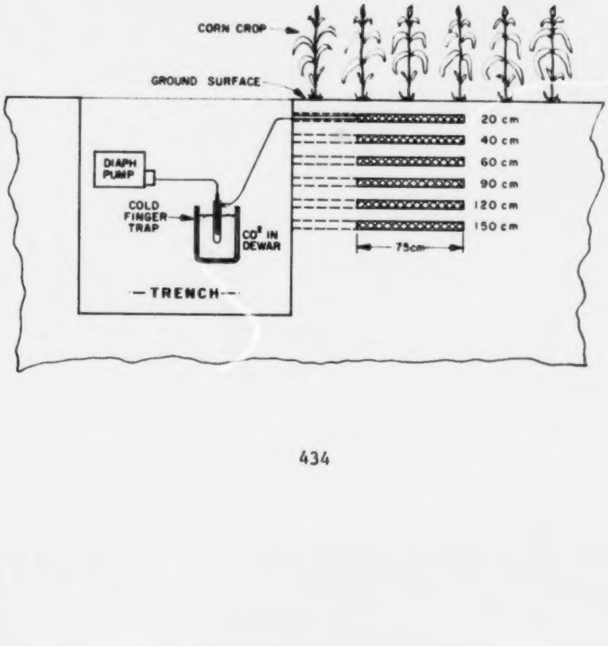


Kelly and Booth [17]developed a model for predicting the movement of tritium to man from an initial wet deposition of tritiated water onto the soil. They compared the results predicted by the model with existing experimental data and found qualitative agreement. Kelly and Booth concluded that with rainfall rates equal to or less than 0.1 cm/d, diffusion becomes important in describing tritium losses from the soil system. At water inputs greater than 0.1 cm/d, translational movement or leaching of the tritium pulse becomes the dominant process determining the half-time of tritium in the soil system. The authors concluded that when conservative input parameters pertaining to soil characteristics, diffusion of soil water, and intake of water by man are assumed, the major factor affecting the infinite dose received by man from a given deposition of tritium on the soil is the rainfall rate.

The movement of tritium in a soil system was studied at the Lawrence Livermore Laboratory (LLL) during the past year. Tritiated water was applied to a young corn crop in a warm, central California valley, which in the summer is a high evapo-transpiration environment. The crop was irrigated daily at approximately 10 to 15 mm water per day after the tritium application. Evapo-transpiration measured by open pan methods indicated that between 5 and 10 mm of water/d were lost in this environment. The soil was a silty clay to a depth of 60 cm, after which the clay content increased. A total of 130 mCi of tritiated water was applied to the 6.2 meter^2 experimental plot, which was within a larger plot of corn to eliminate edge effects. A series of soil vapor lysimeters were installed beneath the corn plot at eight depths up to 150 cm, to sample soil water moving in the profile. Figure 2 is a diagram of the soil water vapor samplers and the experimental plot. The soil water is sampled by pulling the air from the sampler chamber through a cold-finger trap (CO_2) with a small vacuum pump and exhausting the air. The information obtained by this method describes the tritium concentration of the soil water at the installed depth, but yields no quantitative information on the amount of soil water present.

Figure 2

Soil Water Vapor Samplers and Experimental Corn Plot

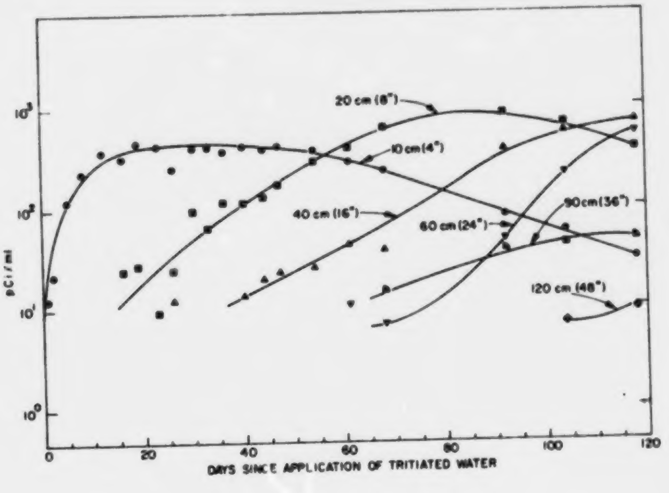


The movement of tritiated water in the soil beneath the corn plot is shown in Figure 3. A broad peak is observed in the 10 cm depth stratum and occurred at this depth throughout most of the growing season. Tritium peaks occurred in successively deeper strata with the passage of time and continued irrigation. The irrigation rate was just slightly above that estimated to be needed for growth and evapo-transpirational requirements and, thus, there was only a small amount of excess soil water to move the tritium pulse deeper into the soil. The tritium peak in the 20 cm stratum occurred 85 days after the tritium application. It is possible that the high water requirements of the corn effectively held the tritium pulse in the surface 0 to 10 cm stratum during the maximum growth period. Irrigated corn with a high biomass production may release from 1.3 to 7.6 mm water per day [18].

The tritium concentrations in soil water from the 20 cm stratum, Figure 3, were higher than in the 10 cm stratum in this series of measurements. This may be explained by the fact that the two samplers (10 cm and 20 cm) were not in a vertical series and the data obtained from each does not represent a linear depth relationship. The 10 cm stratum above the 20 cm sampler has higher concentrations of tritium than the 10 cm stratum measured.

Figure 3

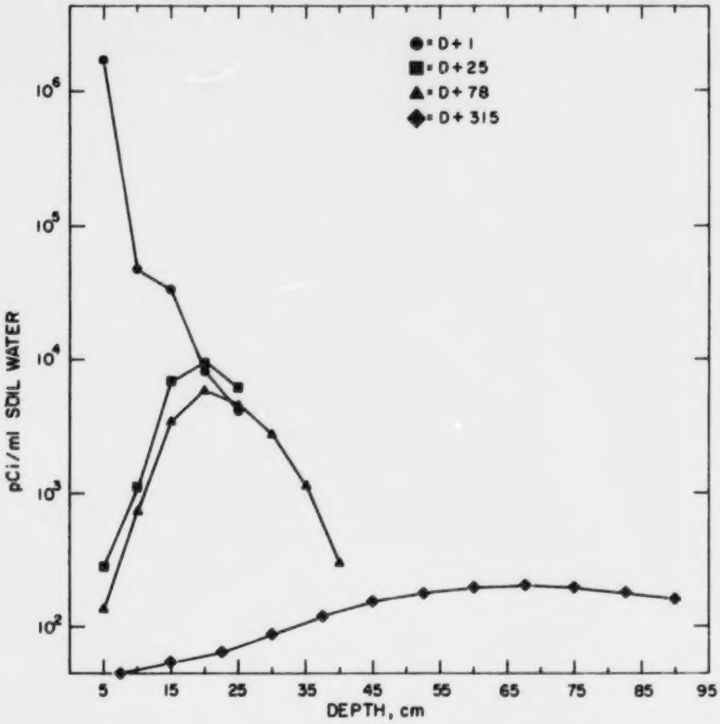
The Movement of Tritium in Soil Water Beneath the Corn Plot



In the spring following tritium application (1971), peak activity was seen at the 40 and 60 cm depths 274 days after application, and tritium concentrations in the surface 20 cm of the soil profile were in the range of 2.2 X 10^2 pCi/ml of soil water. Irrigation of the plot ceased 29 days after harvest and during the winter of 1970-1971, 39.5 cm of rainfall were received. At 350 days post-tritium application, a soil tritium profile was determined from core samples. Data obtained by core sampling at day 1, 25, 78, and 315 post-tritium application are shown in Figure 4.

Figure 4

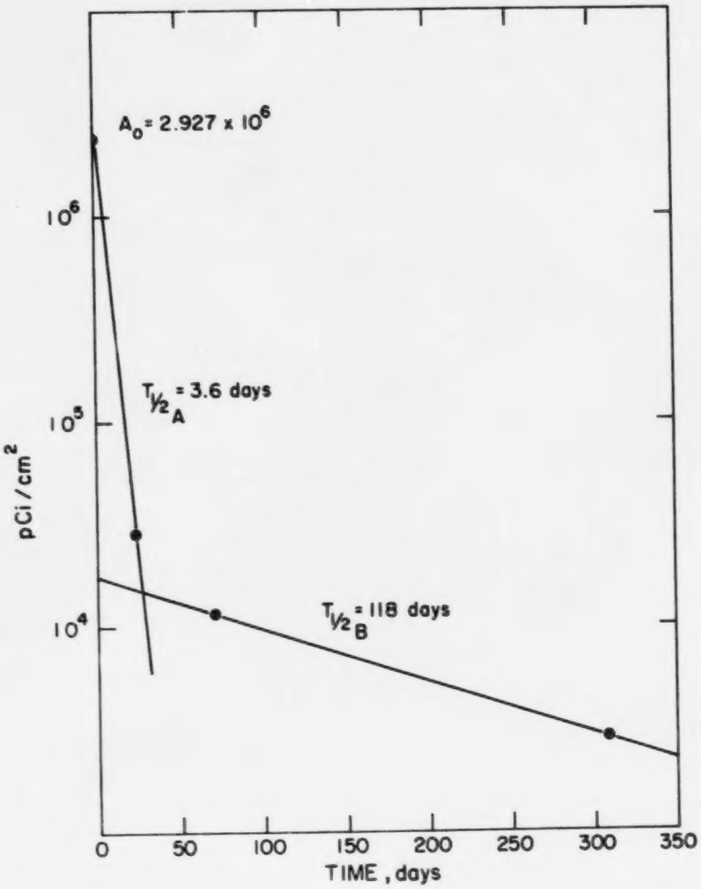
Soil Water Tritium Concentrations Beneath Corn Plot Determined from Core Samples



If the soil tritium distribution profiles in Figure 4, obtained by coring the experimental plot, are integrated and the data expressed as pCi/cm^2 of soil surface, a short and long term decay component are apparent. These data are shown in Figure 5 and indicate a short term component with a half-life of three to six days and a longer component with a half-life of 118 days.

Figure 5

Integrated Soil Tritium Activity and Half-life in Corn Plot



The deposition of tritium as tritiated water vapor on or in soil has not been studied in detail, if at all. The basic phenomenon in this type of exposure is very likely an exchange of tritiated water with stable water in the surface strata of the soil. Experiments are presently in progress at our laboratory to describe tritiated water vapor deposition on and in soils.

The deposition of tritiated water vapor on bare soil was studied in an exposure of a recently cultivated field. The study plot had been plowed and disced earlier in the season. One week before the experiment the soil was compacted with a Brillion compactor in preparation for planting various crops. The soil moisture below the 5 cm depth ranged from 14 to 19% and was derived from rainfall received the previous winter.

The soil was exposed for a period of 50 minutes with a lucite chamber. Subsequent to exposure, the soil was sampled by a trowel and pit method in which a small vertical-walled sample hole was dug, and then depth increments were lifted carefully from the vertical face of the sample hole. The entire profile sampled was involved in the soil preparation previously described, and this may account for the presence of tritium at a depth of 15 cm from the relatively short exposure.

A rapid loss of tritium occurred post-exposure in the 0 to 7.5 cm stratum due to water vapor losses during the hottest part of the day, which occurred just after exposure. On the second day after exposure, 18 to 25 hours post-exposure, tritium loss continued in the surface stratum (0 to 2.5 cm), but in the strata beneath the 2.5 cm depth, redistribution of tritium took place and changes in concentration occurred in all strata with, however, only small changes occurring in the water content of the strata. Diffusion of tritiated water was undoubtedly occurring in the 5 to 15 cm stratum with its higher water content (15 to 19%) while vapor losses or transfer took place mainly in the drier surface stratum (0 to 2.5 cm). This experiment was a preliminary effort concerned primarily with developing techniques appropriate for studying tritiated water vapor uptake by soil.

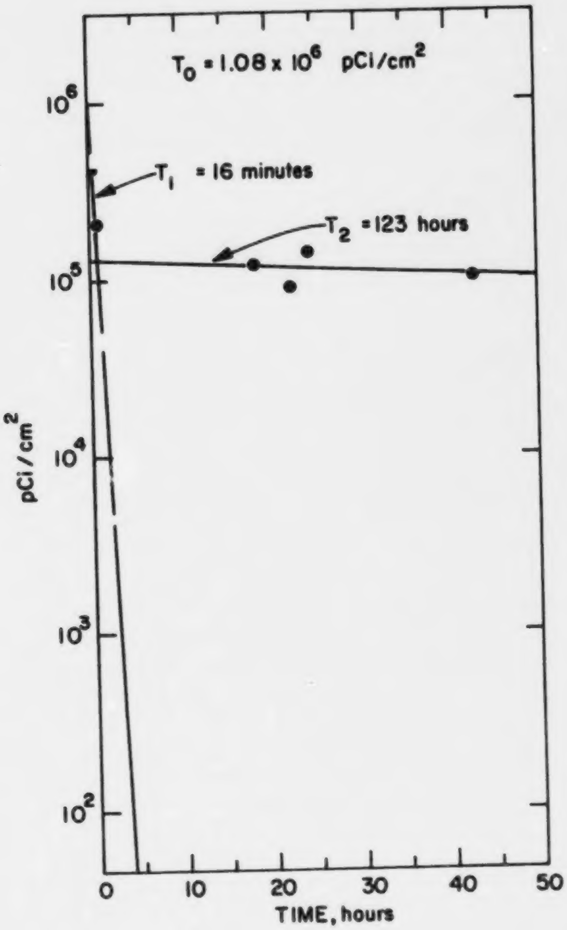
If each tritium depth profile obtained in this experiment is integrated and the values converted to pCi/ml with the appropriate data (pCi/g of soil and g soil/ml), the amount of tritium present in the soil system at each point in time may be calculated. These data are shown in Figure 6 with the half-time of each component of the decay indicated. A rapid initial loss of tritium from the soil profile occurred with a half-time of 16 minutes, as the higher concentrations of tritium evaporated from the surface stratum of soil. Subsequent decay of tritium in the exposed soil took place at a much slower rate with a half-time of 123 hours.

A number of distinct principles emerge from these data on tritium movement in soils. These are:

1. Tritium introduced at the soil surface moves through the soil system at a rate proportional to the amount of water applied or received after tritium exposure; also tritium movement is strongly affected by the mechanical characteristics of the soil.
2. The half-time of tritium in the soil is generally longer than in other components of the ecological system; and the relatively longer half-time in the soil extends the half-time of tritium in vegetation rooted in that soil.

Figure 6

Integrated Soil Water Tritium Profiles After a Single Tritiated Water Vapor Exposure



3. There is an indication that high water use plants, such as crops, may affect the movement of a tritium pulse in the soil.

4. Soil tritium half-times may exhibit short and long components; the short component is tentatively attributed to bulk water movements while the longer component is very likely due to tritium retained in more stationary water, such as chemically bound water.

5. It is apparent that the movement of tritium in the soil will be a critical factor in tritium releases into ecological systems because of long retention times and the availability of the tritium pulse to biological systems during that period.

The movement of ground water containing tritium is a distinct subject from soil water movements and will not be discussed here.

Tritium in plants

Because of the intimate relationship between the soil, soil water, and vegetation growing in an environment, it is difficult to separate completely the subjects of plant and soil water movements. Water in any ecological system exists in a continuum among the soil-plant-atmosphere. Hydrological studies indicate that between 23 and 69% of the incoming precipitation in grassland areas [19] is soon recycled, via this water continuum, back to the atmosphere in the form of evapo-transpiration. If a labeled pulse of water (tritiated water) is injected into this environment, it moves along the same continuum and is partitioned into the various water compartments of that ecological system.

When tritiated water falls on the vegetation it will be absorbed by the leaves and stems of the plant. Tritiated water may enter the plant leaf through the cuticle and also via the stomata. Once within the leaf, it will (1) be involved in the transpirational stream and be swept out of the plant with the large flux of water transpired each day, or (2) it will be involved in photosynthesis and fixed in organic compounds which may be translocated to other parts of the plant.

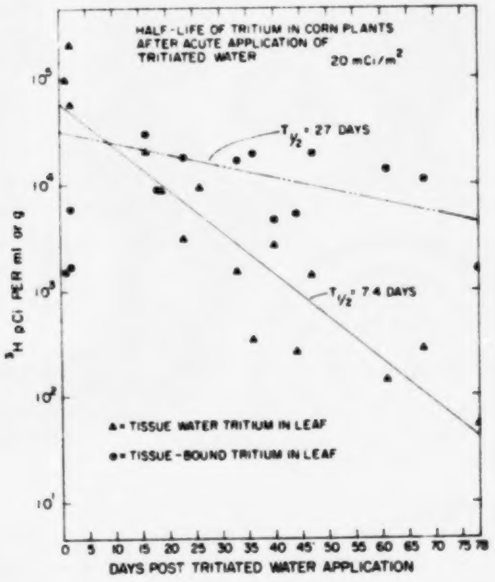
Another compartment in plant tissues which should be considered is the retention of tritium in the free space of the leaves, that is, in the water within and on the cell walls. There is some evidence to indicate that the bulk of water transpired by plants moves through the cell walls rather than through the protoplasm of the cells of the leaf [20].

Vaadia and Waisel [21] studied water absorption by the aerial organs of plants by using tritium, both liquid and vapor exposures. Only time-zero values in the plant leaf tissues were presented in their data. Allepo pine (*Pinus halepensis*) and the sunflower (*Helianthus annua* var. advance) were used in their experiments. When sunflower leaves were exposed for three hours to tritiated water, their tissue-water tritium concentration was 80% of the applied tritium. In pine needles, only 50% of the tritiated water was found in the needle tissue-water in the three hour period. Water-stressed leaves did not acquire as high a concentration as the normally hydrated leaves, especially in the pine. In a one hour exposure, concentrations of leaf-tissue water were approximately 20% of the applied tritiated water in the pine and 60% in the sunflower leaves, both normally hydrated.

In ecological studies conducted at LLL, the exposure of growing corn to a single tritiated water application was made to study both soil and plant behavior. Tritiated water was applied to the experimental plot by hand with a 7.6-liter pressurized garden spray unit. Approximately 1850 ml of tritiated water were applied to a 6.2 m^2 plot of growing corn. The total amount of tritium applied was 139.23 mCi or approximately 22.5 mCi per meter^2. The specific activity of the tritiated water applied was 75.2 μCi/ml. Soil data from this experiment were discussed previously. Plant uptake of tritiated water continued throughout the growing season from the single application of tritiated water because of the long half-time of tritium in the soil. At the end of the growing season, the corn was harvested, and aliquots taken for tritium analysis of tissue-water and tissue-bound tritium. The dried residues of the corn leaves, stalks, ears, and roots were combusted, and the concentration of tissue-bound tritium determined. Tissue-water concentrations of tritium in corn leaves collected during the growing season are shown in Figure 7. Because of the rapid turnover of tissue-water in the corn, with its high transpiration rate, it is very likely that the tissue-water of the leaves is in equilibrium with the soil-water tritium. This has been observed at Sedan crater in the Nevada desert where plants are growing in soil contaminated by tritium [16].

Figure 7

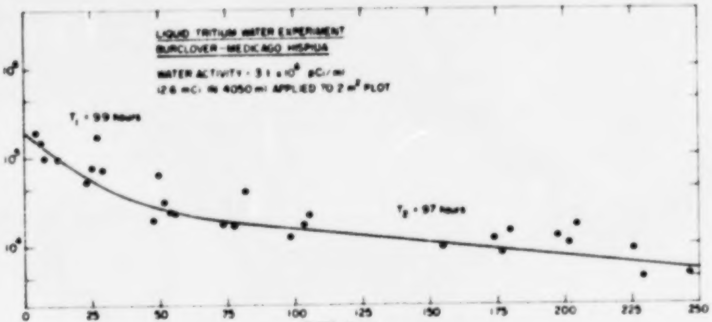
Tissue-water Tritium Concentrations in Corn Plants After a Single Tritiated Water Exposure



In another experiment, the persistence of a single, liquid exposure of tritium was studied in native weedy vegetation which was not irrigated. The burclover, Medicago hispida, was exposed to tritiated water and tritiated water vapor in this series of experiments. Data obtained in the tritiated water experiment are shown in Figure 8. Two components are evident in the decay of tritium in the tissue-water of the burclover after a single tritiated water application. The plants were sampled as soon as the tritiated water had dried on the foliage, three hours post-application. The first component decayed with a half-time of 9.9 hours while the second component half-time was 97 hours. At approximately 80 hours post-exposure, the plants exposed to tritiated water had 1/10 of the initial tritium concentration in their tissue-water. This is in general agreement with observations made by Kirchmann *et al.* [22].

Figure 8

Tissue-water Tritium Concentrations in Medicago hispida After a Single Tritiated Water Exposure



The long-lived component of the tritium decay in the plants exposed to tritiated water is strongly affected by the uptake of tritium from the soil, which will constitute a source of tritium for the plant until it has been dissipated by evapo-transpiration or eluted from the root zone. These plots were not watered during the course of the experiment and the plant continued to absorb tritium from the soil; evapo-transpiration constituted a major loss mechanism for tritium in the late stages of this experiment.

Kirchmann *et al.* [22] studied the incorporation of tritiated water applied as a spray on pasture vegetation and the effects of different seasons upon the pattern of contamination. Their plant uptake data may be compared to those obtained in our tritiated water exposure of corn plants. The data will be discussed more completely in the section on ecological systems.

Tritiated water vapor

The uptake of tritiated water vapor by plant foliage has been studied in the laboratory, and a few studies have been conducted in the field. Vaadia and Waisel [21] exposed sunflower plants (Helianthus annua) and Allepo pine (Pinus halepensis) to tritiated water vapor for varying time periods, up to three hours. Their data describe the time-zero conditions immediately after exposure; the experiments were not concerned with the fate of tritium in the plant. Sunflowers exhibited a higher level of absorption than the pine, both of which were pot-grown specimens. Approximately 10 to 12% of the air concentration of tritium in the water vapor was found in the sunflower leaves after the maximum exposure of three hours, while in the pine, less than 5% occurred in the needles after the same exposure time.

A series of ecological experiments, in which field-grown plants were exposed to tritiated water vapor, were conducted at our laboratory to obtain more detailed information on the retention of tritium after water vapor exposures. Sunflowers (Helianthus annua), oats (Avena sativa), corn (Zea mays), and black pine (Pinus thunbergii) were the plant species involved in these experiments. All of the experimental plants were growing in the field before, during, and after the exposures, and thus were affected by typical environmental factors, such as temperature, sunlight, wind, and humidity. Lucite chambers were utilized to expose sunflowers, corn, and oats while the black pine was exposed with a flexible polyethylene bag.

When the upper portion of the sunflower stem was exposed to tritiated water vapor for one-half hour, the tissue-water of the leaves of the plant attained 49.6 and 44.0% of the tritium concentration in the water vapor. Figures 9 and 10 show the data obtained in two experiments with sunflowers, one of which terminated in seven hours, and the other which extended into the second day. The half-time

Figure 9

Tissue-water and Tissue-bound Tritium Concentrations in Sunflower, Helianthus annua, After a One Hour Exposure to Tritiated Water Vapor

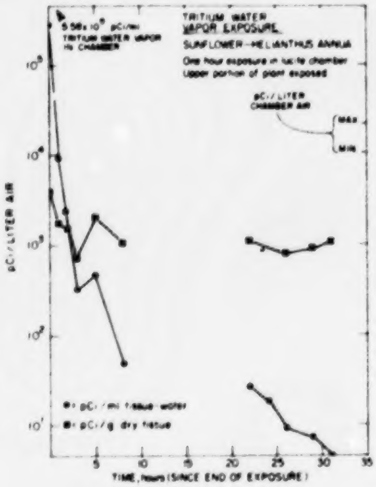


Figure 10

Tissue-water and Tissue-bound Tritium Concentrations in Sunflower, Helianthus annua, After a One-Half Hour Exposure to Tritiated Water Vapor

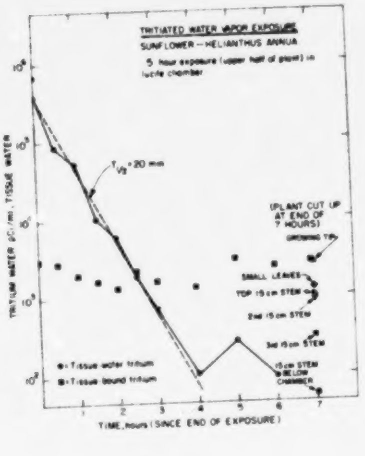
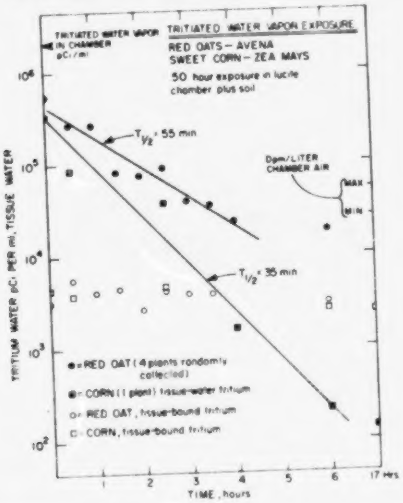


Figure 11

Tissue-water and Tissue-bound Tritium Concentrations in Corn and Oats After a One-Half Hour Exposure to Tritiated Water Vapor



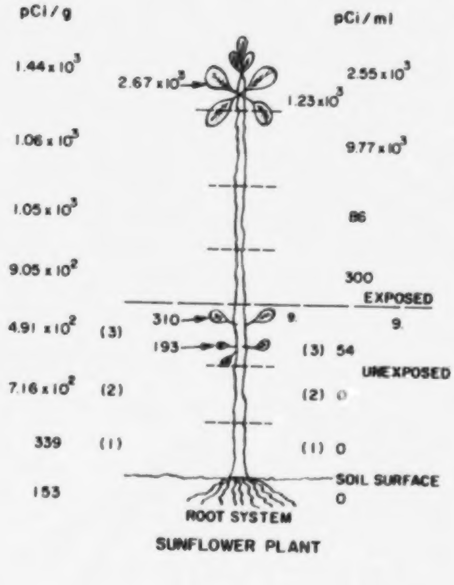
of tritium in the tissue-water of an herbaceous plant like the sunflower under field conditions is approximately 20 to 30 minutes. When red oats are exposed in the same manner, the half-time is somewhat longer, 55 minutes. Figure 11 contains data on the exposure of corn and red oats in the same plot. The corn plants were between 30 and 37.5 cm tall and actively growing, and the half-time of tritium in these plants was 35 minutes.

The leaf and stem samples from the previously described experiments which were analyzed for tritium in the tissue-water were also analyzed for tissue-bound tritium by combusting the lyophilized residues. Data are expressed in pCi/g dry tissue and are shown on Figures 9, 10, and 11 with the tissue-water data. At three to four hours post-exposure in sunflower and corn plants, the concentration of tissue-bound tritium exceeds that in the tissue-water of the sample. The half-time of tritium in this tissue-bound compartment cannot be resolved with any degree of certainty in these experiments because of their short duration.

In the short-term sunflower experiment, terminated at seven hours post-exposure, the entire plant was sectioned at the termination of the experiment and the sections analyzed for tissue-water and tissue-bound tritium. These data are shown in Figure 12. Below the exposed-unexposed interface, there was little tritium detected in the tissue-water. When the same stem sections were analyzed for tissue-bound tritium, concentrations of tritium in the range of 1 M to 1000 pCi/g were found in stem sections which had negligible tissue-water tritium.

Figure 12

Tissue-water and Tissue-bound Tritium Concentrations in a Sunflower, Helianthus annua, Stem Seven Hours After a One-Half Hour Exposure to Tritiated Water Vapor



Apparently, this tritium had been translocated to the stem section as photosynthate synthesized during and shortly after the exposure. Aronoff and Choi [23] and Choi and Aronoff [24] describe the movement of tritiated water and tritium-labeled photosynthate in soybean plants after a one hour exposure to tritiated water. Little tritiated water was translocated from the exposed leaves, but many photosynthetic products were labeled during the exposure. The significance of our data on the sunflower is that tritium-labeled organic compounds will be formed in an exposure as short as 30 minutes, and translocated to other parts of the plant such as stems or roots where they are stored. It is reasonable to propose that with chronic vapor releases of tritium, crop plants could acquire concentrations of tritium in their tissues that would represent in some manner the time-integrated exposures.

The uptake of tritiated water vapor by the black pine, the only woody plant in this series of experiments, was less than in the herbaceous species. The half-time of tritium in the tissue-water of the needles was longer than in other plants studied, Figure 13. Vapor

Figure 13

Tissue-water and Tissue-bound Tritium Concentrations in Black Pine, Pinus thunbergii, Needles After a One Hour Exposure to Tritiated Water Vapor

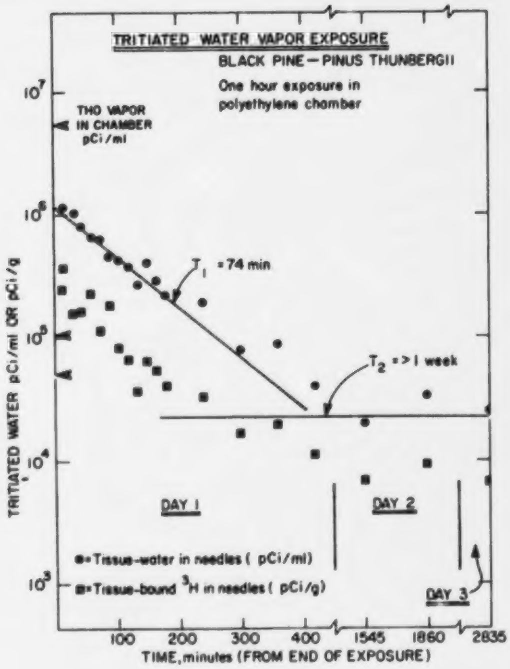


Table 1

Plant Uptake Coefficients for Tritiated Water Vapor

$$\text{Uptake Coefficient} = \frac{\text{pCi/ml tritium in tissue-water of plant at } T_0}{\text{pCi/ml tritium in vapor of exposure chamber}}$$

Plant	Exposure Time (min)	Uptake Coefficient
Black Pine *Pinus thunbergii*	60	0.17
Oats *Avena sativa*	30	0.19 0.21
Burclover *Medicago hispida*	30 60	0.26 0.23
Corn *Zea mays*	30	0.27
Cranebill *Erodium cicutarium*	60	0.29
Wild Barley *Hordeum californicum*	60	0.39
Fiddleneck *Amsinckia grandiflora*	30 60	0.45 0.42
Sunflower *Helianthus annua*	30	0.49

uptake coefficients for eight plant species used in these experiments are shown in Table 1. The order of the species in this table is essentially from xeric to hydric, that is, the plant with the highest rate of water metabolism shows the greatest rate of uptake of tritiated water vapor, and has the shortest half-time of tritium in its foliage. The uptake of tritiated water vapor was highest in the sunflower and lowest in the black pine. The half-time of tritium in the needles of the black pine exposed to tritiated water vapor was 70 minutes, but a second component was suggested by the data which, however, did not extend far enough to resolve the half-time of the longer component.

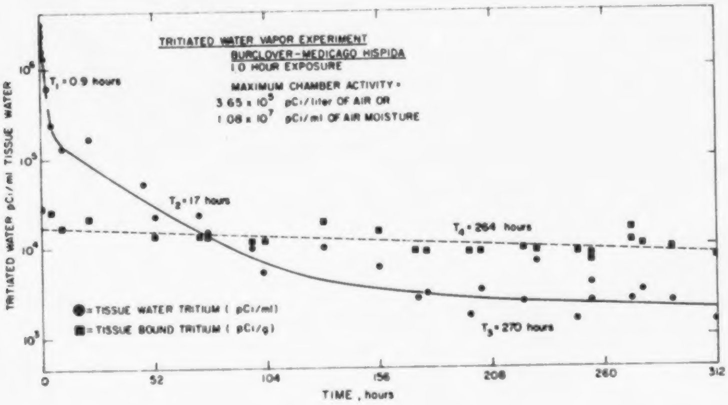
Many installations releasing tritium to the environment are in areas where the natural vegetation is coniferous or deciduous trees. The tissue-water of the leaves or needles of these trees will be in a state of quasi-equilibrium with the tritiated water in the air, and will provide an easily obtained sample material which will describe the ambient and recent tritiated water concentrations in the air.

A study of the long-term effects of a single tritiated water vapor exposure of a field grown plant was made with a one hour exposure of the burclover, *Medicago hispida*, which grows as a weed in central California pastures and fields. A 58 X 58 cm plot of the burclover vegetation was exposed in the 205-liter lucite chamber using a large tritiated water bubbler to create a tritiated water vapor atmosphere in the chamber. The tissue-water and tissue-bound concentrations of tritium in aliquots of the exposed foliage are shown in Figure 14, with related data on the conditions of the exposure.

A three-component decay of tritium activity is seen in the tissue-water of the plant foliage. More than 90% of the initial tritium concentrations in the tissue-water of the leaves is released in the first component which is a half-life of 0.9 hours. The second component accounts for 9% of the absorbed tritium and has a half-time of 17 hours. This component is very likely related to the general water metabolism of the plant, or possibly a biochemical compartment, while the short-lived component (0.9 h) is the loss of tritiated water from the stomatal chambers within the leaves from which it is being flushed by the transpirational stream.

Figure 14

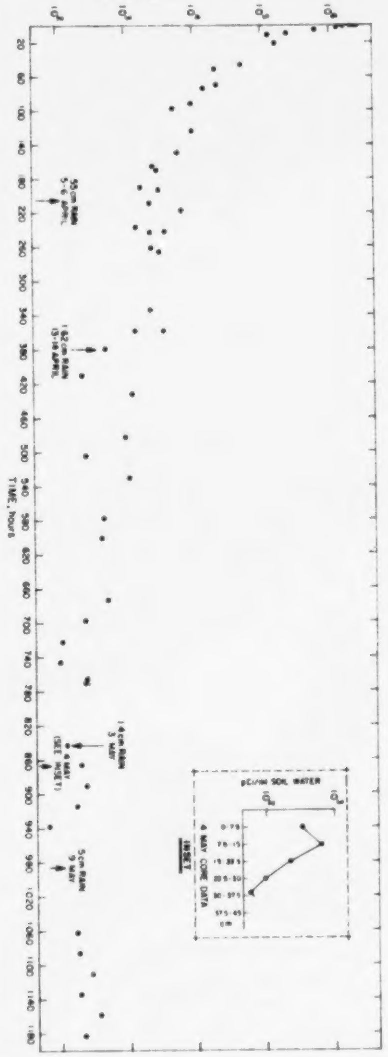
Tissue-water and Tissue-bound Tritium Concentrations in Burclover, Medicago Hispada, After a One Hour Exposure of Tritiated Water Vapor



The third long-lived component of tritium in the foliage of the plant has a half-time of 270 hours and accounts for 0.2% of the initially absorbed tritium. It is attributed to organically fixed tritium in the plant which may be translocated to the roots or used in the metabolism of the stem. Some evidence for this is indicated by the decay of incorporated tritium, which is parallel to this third component in the tissue-water of the plant. Another explanation for the third, long component of tritium in the tissue-water of the leaves is that it represents continued low level uptake of tritium from the soil. Evidence for this is seen in the soil core data obtained 866 hours post-exposure and shown in Figure 15 with other pertinent data on the long component of tritium decay in the plant. Most operational monitoring

Figure 15

Tissue-water Tritium Concentrations in Burclover, Medicago hispida, After a One Hour Exposure to Tritiated Water Vapor - Long Term Study



programs would never observe the short component of the plant tritium half-life, and the second and third components would only be obvious if sampling intervals were frequent.

In any exposure of vegetation to tritium, either as a vapor or a liquid, a variable amount of the tritium exposure will be fixed. The actual rate of fixation is, in general, related to the rate of biomass synthesis. In rapidly growing herbaceous crops, the amount of tritium entering this compartment will be much higher than that being fixed by mature forest vegetation. Only a few ecological studies addressing themselves to the partitioning of a tritium pulse in an entire ecosystem have been conducted. Also, few crop plant studies of tritium uptake under field conditions have been conducted and these data are needed for critical evaluation of many nuclear projects and operations.

Ecological systems

The most useful experiment in determining the movement of tritium in an ecological system is one in which the system is treated as a single unit, with measurements being made at various levels within the system during the decay or movement of tritium. A study of this type was conducted by Jordan *et al*. [12] and Martin *et al*. [25] in the tropical rainforest of Puerto Rico. Tritiated water was applied to the forest floor and its rate of disappearance was determined in the soil water, in the air, and in the leaf tissue-water of trees growing in the plot. These data demonstrate the coupling of ecological compartments with respect to water movement, and show the partitioning of the tritium pulse within the ecosystem at various times after exposure. Very little incorporation or fixation of tritium took place in mature tropical vegetation. However, when young successional species were exposed, measurable incorporation of tritium occurred.

In radioecological studies conducted at Sedan crater, where an entire ecosystem is contaminated by tritium, the uptake of tritium by plants and animals has been studied for six years [16]. In Russian thistle plants (*Salsola kali*) grown to maturity at Sedan crater, equilibrium is nearly achieved between the specific activity of tritium (pCi tritium/g hydrogen) in the tissue-water of the plant and in the tissue-bound tritium in a single growing season in this annual plant. Chorney and Scully [26] also demonstrated this equilibrium condition in bean plants grown in growth chambers.

Koranda and Martin [15] describe tissue-bound concentrations of tritium in the organs of small mammals living at Sedan crater. The animals have been exposed for several generations to tritium incorporated in their foods as well as tritiated water vapor in the soil air of their burrows. Values slightly greater than unity have been observed in the ratio of tissue-bound tritium to body water tritium on a gram hydrogen basis. It appears that this higher specific activity of tritium in the tissues is due to the more rapid decay of tritium in the body water after birth while the tissue-bound tritium in the structural components of the organs decays with a longer half-life. Martin and Koranda [16] demonstrated that the body water of this population of mammals was in equilibrium with tritium concentrations in the soil.

The corporation of tritium into an agricultural crop after a single acute exposure of tritiated water was studied recently at our laboratory. A young corn crop was sprayed with tritiated water (75.2 μCi/ml) fifty days after germination. This experiment was described previously. The crop was irrigated throughout the growing season and harvested at 120 days maturity or 78 days post-exposure. Aliquots were taken of leaves, stems, ears, and roots for tritium assay, and all plant biomass produced was weighed. Tissue-water tritium concentrations in these plants were discussed previously and shown in Figure 5. Tissue-bound tritium concentrations in the harvested corn plants are shown in Table 2. Fixation of tritium in corn ears, which had not been formed at the time of tritium application, is on the order of 500 pCi/g which is closely related to the concentration of tritium in the soil water during the growing season. This level of fixation occurred with daily watering with stable water.

Table 2

Total Tritium Activity and Fraction as Tissue-Bound Tritium in Harvested Corn Plants*

(Data are expressed in μCi)

Plant	Total Tritium in plant at harvest	Total Tritium in tissue-water at harvest	Total Tritium tissue-bound at harvest	% of Total Tritium as tissue-bound	% of Total Tritium in ears at harvest
CO-1	0.51	0.02	0.49	95.5	25.2
CO-3	0.48	0.04	0.43	90.7	15.9
CO-4	1.75	0.60	1.14	65.0	13.5
CO-6	0.52	0.06	0.46	88.6	14.6
CO-9	1.02	0.08	0.94	92.1	31.4
CO-15	1.65	0.36	1.29	77.9	34.6
CO-12	1.76	0.11	1.66	94.0	29.9
CO-36	1.24	0.17	1.07	86.9	11.9
CO-27	1.35	0.09	1.26	94.0	30.0
CO-54	2.84	0.13	2.71	95.9	17.9

*These ten plants demonstrate the range of tritium activity found in the eighty plants grown on the experiment.

The long half-time of the tritium in the soil profile provided the plant with labeled water during its entire growth period. A material balance calculation for the entire ecological system and the tritiated water applied to the corn crop is shown in Table 3.

The level of tritium fixation achieved after a single exposure of a common crop plant to tritiated water, and the maturation of that crop under typical agricultural conditions suggests that as tritium concentrations in the biosphere increase, it may be advisable to develop maximum permissible concentrations for tritium in an organic form. The relatively rapid flux of body water through the mammalian body, however, would remove this form of tritium once it had been catabolized. On the other hand, organified tritium would have a different fate in the mammal, and body water kinetics may play a secondary role in its loss and excretion. This type of tritium intake conceivably could originate in vegetables and fruits grown in another region from which the person lives and obtains his water intake, but where tritium is released to the agricultural environment.

Table 3

Preliminary Material Balance
Single Exposure of Corn Plot to Tritiated Water

	μCi of Tritium		
	Measured	Calculated	Remainder
Tritium applied to corn crop	1.39×10^5		
Total tritium in crop at harvest	1.51×10^3		
Tritium residual in soil at harvest	9.01×10^2		
Tritium lost during growth of crop			1.37×10^5
Tritium loss by transpiration of crop		1.38×10^4*	
Tritium loss by evaporation from soil		1.23×10^5	

Fraction of applied tritium in crop at harvest	1.1%
Fraction of applied tritium in soil at harvest	0.6%
Fraction of applied tritium transpired by corn	9.9%
Fraction of applied tritium evaporated from soil	88.4%
Fraction of applied tritium lost by evapo-transpiration.	98.3%

*By assuming the water volume of shoot transpired each day.

Kirchmann *et al*. [22] studied the incorporation of tritiated water applied as a spray on pasture vegetation and the effects of different seasons on the pattern of contamination. They measured the transfer of tritium in forage vegetation to cow's milk. Levels of tritium found in the organic constituents of the milk from the ingestion of tritium-contaminated forage were compared to those observed when the cow drinks either single or repeated doses of tritiated water. Their plant uptake data may be compared to those obtained in our corn plot experiment which received a single exposure of tritated water. At harvest, our corn plants showed the same pattern of tritium distribution between the tissue-water and the dry matter of the plant; higher concentrations of tritium were found in the dry matter than in the tissue-water at harvest. Kirchmann *et al*. found that 99% of the deposited activity on pasture vegetation was lost in the first day after application. Higher levels of organically-bound tritium in the milk were observed when cows ate tritium-contaminated forage than when they drank tritiated water.

Conclusions

Tritium movement in ecological systems is complex and involves many compartments, both physical and biological. The uptake of tritiated water and vapor occurs rapidly in plants and animals, but because of the rapid turnover of body or tissue-water in living organisms, the half-times are generally short. Plant half-times may be measured in minutes or hours, while animal body-water half-times are one to two days to 15 days. In plants, organic fixation of tritium occurs even in exposures of short duration and the half-time of tissue-bound tritium is considerably longer than that in tissue-water.

Tritium movement in soils is affected by soil characteristics such as bulk density and the displacement of the tritium pulse within the soil system by subsequent increments of rain or irrigational water. Intermediate to long half-times are observed in soil systems with the major factors affecting loss rates being porosity and rainfall rate. In well-drained, porous soils, vapor losses may be effective in dissipating tritium pulses while in denser soils with higher rainfall rates, leaching of soil water is the dominant factor.

The half-time of tritium in plants growing on tritium-contaminated soil is extended by the longer half-time in the soil. The root zone of the plant will contain tritium for varying lengths of time depending upon rainfall rate and soil characteristics.

The following summary statement concerning the movement of tritium in ecological systems may be made:

1. The behavior of tritium in soil systems exhibits a rapid, initial loss with the remaining activity decaying with intermediate to long half-times; the half-time of tritium in the soil system will determine the effect of the exposure on the rest of the ecological system.
2. The behavior of tritium in plants is similar for vapor or liquid exposures with a large fraction of the deposited activity being lost in a short-lived component of the half-life decay. A large fraction of the remaining activity after the initial, rapid decay may be organically bound in plants exposed to tritiated water.
3. The behavior of tritium in ecological systems will vary with the flux of water through the system and the rate of biosynthesis. Half-times in agricultural systems will be short, but high rates of incorporation will occur.

ACKNOWLEDGMENTS

This work was performed under the auspices of the U.S. Atomic Energy Commission. The assistance of Robert Wikkerink and Marshall Stuart in these studies is gratefully acknowledged.

Reference to a company or product name does not imply approval or recommendation of the product by the University of California or the U.S. Atomic Energy Commission to the exclusion of others that may be suitable.

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TRANSFER OF TRITIUM FROM METHANE TO VEGETATION

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The detonation of the nuclear explosive during the U.S. Atomic Energy Commission's Project Gasbuggy opened the way for utilizing nuclear detonations to develop heretofore inaccesible natural gas resources. Results of analyses of the gas taken from the Gasbuggy cavity indicate that tritium is one of the primary radioactive constituents of the natural gas [1].

The study reported in this paper was designed to obtain information on the transfer of tritium from methane, the primary constituent of the gas, to vegetation. Testing of the production of the Gasbuggy well was done by flaring (burning) the gas for extended periods of time. The primary radioactive byproduct of this operation is tritiated water. In order to provide additional information, a set of treatments similar to those of the methane study was used to give an indication of the transfer of tritium from tritiated water vapor to vegetation.

Methods

Fourteen-month-old alfalfa (Medicago sativa var. Lahontan) and 18-month-old alta fescue (Festuca arudinacea) plants growing in standard greenhouse flats were used in this experiment. The alfalfa was in the log phase of growth following the second cutting and had reached a height of approximately 13 inches. The alta fescue was in the growing period following a first cutting and was approximately 11 inches tall.

For the experimental trials, the flats of plants were placed in a growth chamber designed to provide a self-contained air system. The temperature of the chamber was maintained at 21.6±0.4°C; the relative humidity was controlled at 54.4±1.3%; the windspeed was maintained at 225±10 fpm (as measured by a hot wire anemometer, set at the top of the plants); and the light intensity was approximately 2,500 foot candles at the top of the vegetation.

For each treatment, a flat of alfalfa and a flat of alta fescue were placed in the growth chamber and allowed to adjust, for a period of one hour, to the conditions set by the experiment. At the end of the adjustment period, either tritiated methane or tritiated steam of known tritium content was injected into the chamber. Initial concentration of tritium was estimated to be approximately 1 $\mu Ci/m^3$ of air. The total methane concentration was ten liters of methane/m^3 of chamber air for the trials where this gas was used.

In order to verify the air concentration of tritium, samples of the air were collected in a liquid nitrogen cryogenic sampler. The water fraction was analyzed by liquid scintillation and the methane fraction by combusting the gas sample in a tube furnace, then collecting the water. Analysis was by liquid scintillation.

At the end of preselected time intervals, an exhaust fan was turned on as the chamber door was opened. This induced a flow of clean air into the box and away from the flats of plants which were taken from the chamber at this time. The maximum error in the time of treatment was not greater than one-half of one percent of the total time for the treatment.

At each time period, four replicates of the vegetation were collected and placed in plastic bags which were then sealed. The samples were weighed, then the water was extracted by a vacuum distillation process. Extracted water was analyzed by liquid scintillation. After extraction, the samples were placed in a convection oven and dried for 24 hours at 70°C. The dried plant materials were then ground in a mill and ashed in a tube furnace containing a platinum catalyst. The furnace was purged during ignition with water-free oxygen. Water resulting from combustion was caught in a liquid nitrogen freeze trap, then analyzed by liquid scintillation. This latter analysis gives a measure of the tissue-bound tritium.

Results and Discussion

The measured air concentrations of tritium obtained in this experiment are shown in Table 1.

Table 1

Measured Air Concentrations of Tritium

Time (min)	CH$_3$T Treatment (nCi/m^3)	HTO Treatment (nCi/m^3)
5	11	17
10	47	6.5
20	40	5.6
40	35	5.7

The average depositon velocities for each of the contaminating materials is shown in Table 2.

Table 2

Deposition Velocities for Tritiated
Methane and Tritiated Water Vapor

	Deposition Velocity cm/sec	
Crop	CH$_3$T X 10^{-2}	HTO
Alfalfa	5.5 ± 0.4	1.4 ± 0.3
Alta fescue	5.2 ± 0.2	1.2 ± 0.2

Figures 1 and 2 present data showing the tritium content of both the tissue-bound fraction and the "free"-water fraction of the vegetation.

The conceptual model in Figure 3 can be used to present one possible mechanism that explains the interpretation of the data.

Figure 1

Uptake of Tritium by Alfalfa and Alta Fescue
(Source: Tritiated Methane)

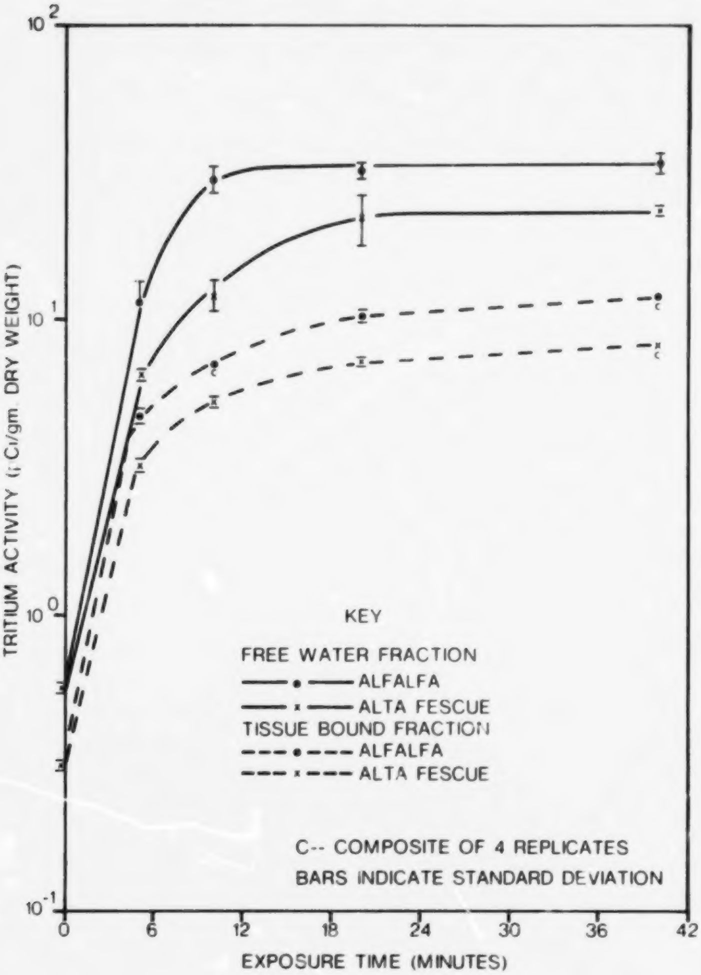


Figure 2

Uptake of Tritium by Alfalfa and Alta Fescue
(Source: Tritiated water vapor)

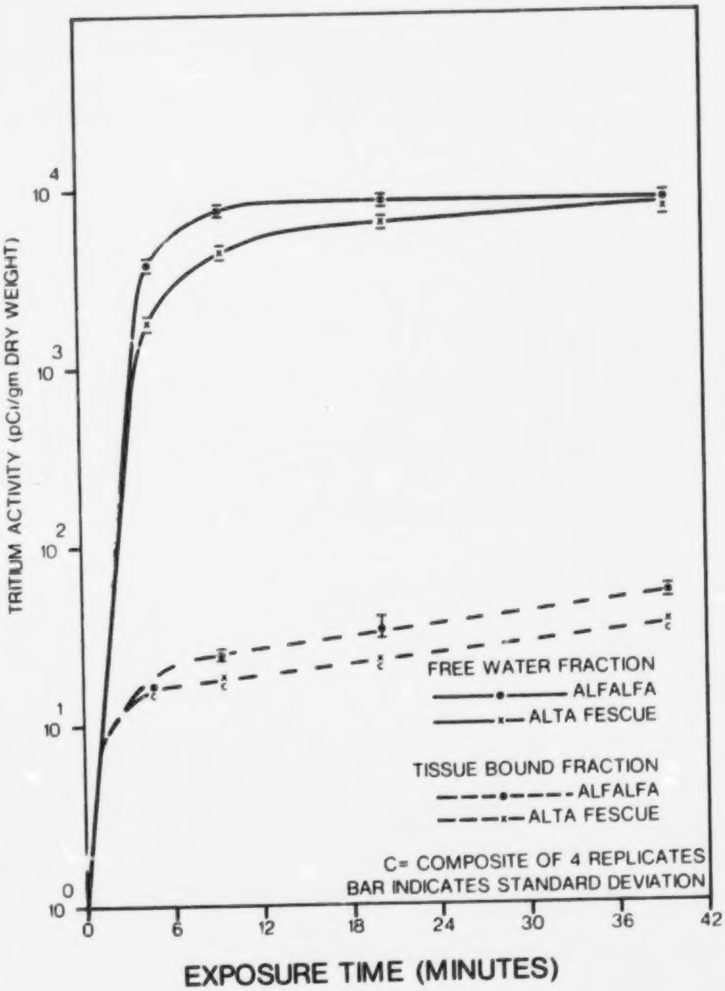
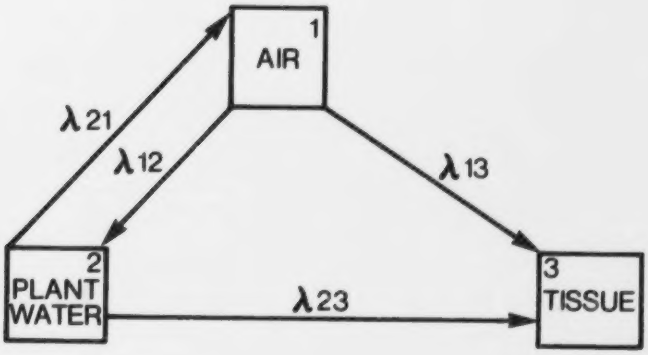


Figure 3

Conceptual Model for Short-Term Tritiated Gas Deposition



The shape of the curves showing the uptake of tritium into the water fraction of the vegetation indicates that the uptake is rapid during the first few minutes of exposure, but an equilibrium is established in a short time due to the turnover of water during transpiration. The difference between the alfalfa and alta fescue is undoubtedly due to this mechanism. Alfalfa is known to be a more active mover of water due to its larger leaf surface area per plant and the larger number of stomates on the leaf surfaces; thus it would be likely to reach equilibrium in a shorter time interval.

The large difference in uptake between the methane and the water vapor is thought to be primarily the result of the much slower rate of tritium exchange in the methane-water system than in the water-water system. Using alfalfa as an example and assuming that equations (1) and (2) control the uptake by the water fraction, it is interesting to note that the ratio between the change in free energy for the two reactions is approximately the same as the ratio between the deposition velocities for the two gases, i.e. 25:1 and 24:1, respectively. Although this observation may be purely coincidental, it seems to reflect a reasonable explanation.

$$CH_3T_{(g)} + H_2O_{(2)} \rightleftharpoons CH_{4(g)} + HTO_{(l)} \qquad (1)$$

$$HTO_{(g)} + H_2O_{(l)} \rightleftharpoons H_2O_{(g)} + HTO_{(l)} \qquad (2)$$

Once the tritium is in the water fraction, it should be available for incorporation into the organic constituents of the tissue. However, work done at this laboratory and elsewhere [2] indicates that this rate is so slow that $\lambda 23$ could not account for the levels seen in Figures 1 and 2 during the 40-minute exposure time. The fact that the uptake of tritium into the tissue fraction is approximately the same for both contaminating materials suggests that a common mechanism is operating to take tritium into the tissue.

It is known that vegetation contains a large surface area. Since both gases are known to react readily with surfaces, especially those containing sites for hydrogen bonding, it is hypothesized that the initial uptake of tritium is via absorption onto the surface of the vegetation. Absorption should occur very rapidly and should have a limit to it. Also, if absorption onto charcoal is any indication, water should be slightly more reactive than methane. The tissue curves in Figures 1 and 2 can be interpreted to result from these mechanisms.

The fact that both levels of activity continue to increase after the initial, rapid rise probably reflects the slower transfer via $\lambda 23$. This observation is supported by the fact that the rate of tritium uptake in this portion of the curve is essentially the same for both gases. If a portion of the tritium was first transferred to water, then to tissue, such a similarity in the rates would be expected.

Attempts to do a mass balance study of the tritium injected into the chamber proved to be impossible. This is believed to be due to the absorption of tritiated gas onto unmeasured components of the system such as the walls of the chamber, the wooden flats, and the soil in the flats.

Conclusions

Although the mechanism of deposition proposed in this paper is speculative, it offers one explanation. Others no doubt exist. The deposition velocities obtained indicate considerable differences between the two sources of tritium contamination. These values should prove helpful in assessing the impact of future natural gas stimulation tests conducted under the Plowshare program.

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THE BEHAVIOR OF TRITIUM IN THE SOIL

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There are many kinds of investigations that require knowledge about the interaction between tracer or pollutant in solution and porous media through which water flow occurs. Soil physicists, engineers, hydrologists and other scientists are interested in predicting the rate of water movement through porous media and, for many kinds of investigations, this can most adequately be accomplished by the use of tracer techniques. Some water tracers are classified as potentially hazardous pollutants and are, therefore, of special interest to environmental engineers, conservationists, ecologists and to all who have a direct interest in water quality. Tritium, a radioactive isotope of hydrogen, is being used extensively as a tracer to depict water flow in porous media and, because it is a product of thermonuclear reactions, it is sometimes considered a pollutant to the environment. Since the advent of the thermonuclear bomb, tritium has been released to the environment in large quantities. Bond [1] estimates that all the tritium produced by nuclear explosions will eventually be converted into tritiated water and mixed with water circulating in the hydrologic cycle. Hydrologists have successfully used bomb-produced rainout tritium as a hydrologic tool to trace underground water.

Although many investigations have been conducted to determine the behavior and fate of tritium in the environment, there is still a lack of knowledge about its behavior in some natural processes, such as its interaction with soil materials. It is not only important to understand the fate of tritium in natural processes, but it is also important to fully understand its potential as a scientific tool. Listed below are reasons for concern about the behavior and interaction of tritium in soils; in fact, these same factors provided the impetus for this investigation.

First, there is a continuing search for an ideal water tracer. For many kinds of investigations, tritium offers numerous advantages over other tracers. It has become one of the most widely used water tracers because: it is incorporated directly in the water molecule; it is radioactive with a sufficiently long half-life for many practical hydrologic investigations; it is inexpensive and easily monitored in tracer concentrations and, therefore, large quantities of water can be spiked inexpensively; and, for practical investigations, its behavior resembles that of protium closely enough so that its movement depicts the movement of water with an accuracy equal to or better than other commonly used tracers.

Second, most applications of the tritium tracer technique require an understanding of the isotope effect, and some require applying isotope effect corrections to experimental data. Therefore, knowledge about the interaction of tritium with mineral surfaces and about the behavior of tritium in the soil water system needs to be clarified before its true potential as a water tracer and its fate in the environment will be fully understood.

Third, the isotope effect phenomenon, when better understood, may provide a good scientific tool to study clay-water interaction and other natural processes occurring in the clay-water system.

The Movement of Tritium in Soil

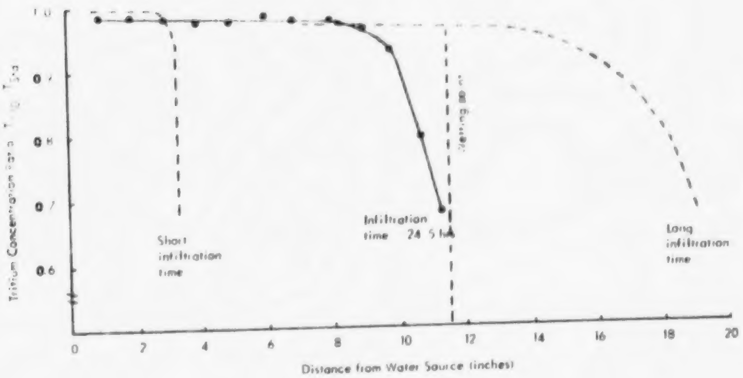
The literature contains what appear to be conflicting statements about the movement of tritium in soils and the degree of its interaction with soil surfaces. Stewart [2] reports that during the diffusion of tritiated and deuterated water in soil columns containing oven dry Davidson clay, the water near the wetting front becomes diluted with these isotopes and the degree of dilution is a function of distance to the water source. His data suggest that there is a retardation in the flow velocity of tritium compared to protium or bulk water flow. Kaufman and Orlob [3] and Biggar and Nielsen [4] found a slight retardation in the flow velocity of tritium compared to chloride flow. Because chloride is an anion it experiences repulsion by negatively-charged mineral surfaces which could result in its diffusion rate being greater than that of bulk water. Corey and Horton [5] studied the movement of deuterium, tritium and oxygen-18 during miscible displacement through acidic kaolinitic soils and found no measurable difference in the relative flow rates. These investigations suggest that, for most practical hydrologic investigations, it is unlikely that isotopic interaction will be great enough to require special corrections or interpretation of experimental data. Other factors related to the complicated nature of flow in stratified soil profiles are more significant and unpredictable; also, isotopic interaction between solid and liquid phases in sands and gravels is insignificant, and the movement of tritium should accurately depict the flow of water through them.

For some investigations where water flow or pollutant transport is being traced through clays, the interaction of tritium with clay surfaces is more important. Factors that are involved and that may explain the apparent differences in experimental findings mentioned above include: type of soil used, the physical condition of the soil and its initial water content, the type of flow studied (saturated vs. unsaturated), and other experimental conditions. It is likely that for some experimental conditions the interaction of tritium with soil surfaces will be different from that of protium, which will result in a difference in flow velocity. The two extreme conditions, oven dry soil and completely saturated soil, are not the conditions that are normally present in surface soils and during water flow therein. Surface soils do become rather dry, and saturated and near-saturated conditions often exist immediately following heavy rains. For a few special conditions, such as in swamp areas, slow water movement occurs under saturated conditions. The water content of surface soils is constantly changing. Therefore, more experimental work is required to understand the behavior of tritium under changing conditions that exist in nature and for different soil types.

The curves in Figure 1 are drawn to show what is expected if significant interaction occurs between tritium in solution and

Figure 1

Tritium concentration ratio (tritium concentration of water released in a furnace at 110°C divided by the tritium concentration of the infiltration water) as a function of distance from the wetting source. The center curve and the location of the wetting front are drawn from data of Stewart (2). The other two curves are drawn to represent graphically what would be expected for short and long infiltration times when some of the tritium becomes immobilized with porous media.



soil surfaces. The solid line is actual data reported by Stewart [2], whereas the dashed lines represent what might be expected during early and long infiltration times. If no interaction occurs, the concentration ratio is equal to one from the water source to the wetting front. However, because of interaction between tritium in solution and soil surfaces, there is a decrease in the concentration ratio as the wetting front is approached. Some of the tritium becomes immobilized as the diffusion of tritium proceeds because of isotopic exchange and interaction. When diffusion is accompanied by adsorption, the diffusion process in one dimension can be described by the equation

$$\frac{\partial C}{\partial t} = D\frac{\partial^2 C}{\partial x^2} - \frac{\partial S}{\partial t} \qquad (1)$$

where C is the concentration of the tracer, t is time, x is distance, S is the amount of immobilized tritium, and D is the diffusion constant. Crank [6] discusses possible solutions to this type of diffusion equation.

The Isotope Effect and the Clay-Water System

Because of the large relative mass difference between the hydrogen isotopes, an isotope effect occurs as water passes from one physical state to another. It may be large or small depending on the nature of the transformation and the energy required to break and reform hydrogen bonds. For example, during simple evaporation, the change in physical state of water results in a small isotope effect where, at equilibrium, the vapor phase is found to contain about 10% less tritium than the liquid phase. Water and/or hydroxyl groups are associated with soils in different physical and chemical states which provide mechanisms for an isotope effect. It is present as: water vapor, water of hydration on cation surfaces, tenaciously adsorbed water on mineral surfaces, interlayer water, and bulk pore water. In addition, hydroxyl groups make up an intimate part of the crystal lattice of clay minerals, and some of these hydroxyls, primarily broken edge and surface exposed hydroxyls, are labile to exchange with tritium in the adsorbed water phase. The behavior of tritium as water is transferred from one of these physical states to another is discussed below in reference to tritium fractionation.

The Vapor Phase

The isotope effect phenomenon is involved in several processes that occur in nature, such as during the evaporation of water where the vapor phase is depleted in tritium compared to the liquid phase. Conversely, condensation from a limited amount of vapor results in an enriched condensate and a remaining vapor depleted of tritium. These phenomena provide the fundamental reasons why deuterium and oxygen-18 concentrations vary appreciably in natural waters. During evaporation, the fractionation factor, α_T, for tritium (or for any of the isotopes incorporated in the water molecule) is equal to the ratio of vapor pressures of the tritium component and the protium component:

$$\alpha_T = \frac{e_{THO}}{e_{H_2O}} \qquad (2)$$

where e_{THO} and e_{H_2O} are the vapor pressures of THO and H_2O.

It can also be expected that equation (2) would describe isotopic fractionation during evaporation from air-water interfaces in the soil and that it can be used to study the influence of clay-water forces on fractionation.

Soil water is continually moving in response to potential energy gradients; measurement of vapor pressure gradients is one method used to measure the potential energy gradient in soils. The potential energy gradient causes soil water to be in continuous movement in both liquid and vapor phases. Movement in the vapor phase is because of evaporation in a wet region followed by condensation in dry soil or cold regions. Because isotopic fractionation occurs during both evaporation and condensation, there may be considerable variation in the tritium concentration of soil water, particularly at the soil surface where maximum evaporation occurs and where water content fluctuations are the greatest.

The Adsorbed Water Phase

Water molecules at a vapor-liquid interface are attracted upward less strongly than downward, which results in a potential energy difference in going from the liquid to the vapor state. Similarly, clay-water forces cause a potential energy gradient in going from bulk pore water to within close proximity of solid surfaces of clay minerals. In unsaturated soils these clay-water forces extend to the vapor-liquid interface and influence the escaping tendency of water molecules from the liquid surface. Vapor pressure is, therefore, a direct measure of the energy status of soil water. Stewart [7] gives the following thermodynamic equation to show the relationship between vapor pressure and matric potential of soil water:

$$\frac{e'}{e} = \exp \frac{v}{RT} (P' - P) \qquad (3)$$

where e' and e are the vapor pressures of adsorbed water at corresponding external pressures P' and P (P' - P is commonly called soil water potential or matric potential), v is the partial molar volume of adsorbed water and is assumed constant, R is the gas constant, and T the absolute temperature. It is generally believed that, because of clay-water forces, there is a high degree of orientation in the first few molecular layers of tenaciously adsorbed water, and that the physical state of this oriented water is different from that of bulk pore water [3,8]. According to van Olphen [9] the first, second, third and fourth molecular layers of tenaciously adsorbed water on montmorillonite surfaces are held at energies of 140, 65, 32 and 5 ergs/cm^2, respectively. A measureable isotope effect may be expected as tritium is exchanged from the physical state of bulk pore water to the physical state of tenaciously adsorbed water.

Stewart [7] combines equations (2) and (3) to relate isotopic fractionation to the matric potential of adsorbed water, thus giving,

$$\alpha'_T = \alpha_T \exp\left[\frac{(v_T - v) \quad (P' - P)}{RT}\right], \qquad (4)$$

where $\alpha_T = e_T/e$ is the fractionation factor at a reference potential P, $\alpha'_T = e'_T/e'$ is the fractionation factor at a matric potential P', and v_T and v are the molar volumes of tritiated water and soil water, respectively.

Equation (4) suggests that very little fractionation of tritium is expected in the adsorbed water phase. Stewart's [7] experimental data also show that fractionation is not measureable between tenaciously adsorbed water and bulk pore water for Davidson clay and the following reference clay minerals: kaolinite, montmorillonite, vermiculite, illite and halloysite. However, his experimental methods did not allow separation of bulk pore water from the first two or three molecular layers of adsorbed water where tritium fractionation would be at a maximum. Tritium fractionation for simple Raleigh type evaporation and condensation may be responsible for greater differences in the tritium concentration of soil water, and perhaps even within the adsorbed water phase itself, than can be explained by clay-water forces alone. Because clay-water forces are partly responsible for vapor pressure gradients in the soil, they are also a factor in evaporation and condensation processes and may, therefore, partly be the cause of tritium concentration differences in the soil.

Crystal Lattice Hydroxyls, Isotopic Exchange and Fractionation

Several investigations have been conducted to study exchange between hydrogen in adsorbed water and hydroxyl groups associated with the crystal lattice of clay minerals [2,10,11,12]. Deuterium has been more widely used than tritium in these investigations. The primary weakness in most of this early work was that little concern was given to the possibility of an isotope effect; the experimental data generally was reported as the percent exchangeable hydroxyls. For some experimental conditions and types of clay minerals, Stewart [2] finds that isotopic fractionation does occur and that it may be significant.

If all crystal lattice hydroxyls are exchangeable with hydrogen in the adsorbed water phase, then the fractionation factor, α_T, could be estimated by the concentration ratio:

$$\alpha_T = \frac{[T]'/[H]'}{[T]/[H]} \quad . \qquad (5)$$

For dilute concentrations where $[T]' << [H]'$, $[T] << [H]$ and $[H]' \approx [H] \approx 100\%$, the ratio becomes

$$\alpha_T = \frac{[T]'}{[T]} \qquad (6)$$

where equilibrium is assumed and $[T]'$, $[H]'$, $[T]$ and $[H]$ are the concentrations of tritium and protium in crystal lattice hydroxyls (primed) and in the associated adsorbed water phase (unprimed), respectively. It is not reasonable to assume that all crystal lattice hydroxyls are exchangeable and, therefore, because of isotopic fractionation, there is no convenient method to determine either α_T or the percent of exchangeable hydroxyls. Concentration ratio measured experimentally and given by equations (5) and (6) may be a combination of both fractionation and the percent exchangeable hydroxyls. The values reported as the percent exchangeable hydroxyls for previous investigations probably represent maximum rather than absolute percentages.

Tritium and deuterium concentration ratios for some common clay minerals that had equilibrated for long periods of time prior to isotopic analysis are given in Table 1. These ratios generally agree with those measured by other investigators, and they were reproducible except for an occasional set of samples which showed considerably more tritium exchange than indicated in Table 1. In fact, some samples showed tritium fractionation of considerable magnitude in crystal lattice hydroxyls. Deuterium concentration ratios for all samples, including those having high ratios for tritium, were about the same as given in Table 1, which suggests that the unique properties of tritium, such as the ionizing effects of tritium β decay, are involved [13]. Tritium concentration ratios greater than one indicate isotopic fractionation, and if only a small percent of the hydroxyls are exchangeable, as believed by most mineralogists, then the isotope effect would be large.

Four samples of Davidson clay, a kaolinitic field soil, gave ratios greater than one and as high as 3.1 after 17 months equilibration. A series of illite samples also gave ratios greater than one with one sample having a ratio of 1.8 after 19 months equilibrium. These data suggest that considerable isotopic exchange occurs between adsorbed water and crystal lattice hydroxyls which is accompanied by a moderate amount of fractionation, or that very little isotopic exchange occurs which is accompanied by considerable fractionation. Attempts to reproduce these data having ratios greater than one have been unsuccessful. Samples giving high exchange and fractionation were subjected to the same pretreatments and experimental conditions as the samples which supplied the data for Table 1. Investigations are under way to determine the reason and factors associated with high exchange and/or large tritium fractionation as shown for some occasional illite and Davidson clay samples.

Summary

Because of the characteristics of the clay-water system, tritium fractionation occurs as tritium in bulk pore water exchanges with water in different physical states and with hydroxyl groups. Both thermodynamic theory and experimental data suggest that tritium fractionation is small as isotopic exchange occurs between the bulk pore water phase and the tenaciously adsorbed water phase. Isotopic exchange occurs between hydrogen in adsorbed water and crystal lattice hydroxyls for all clay minerals and soils studied. A few samples of illite and Davidson clay showed high exchange rates and suggested that considerable tritium fractionation may occur. The conditions and/or factors responsible for this high exchange are not known. Many practical hydrologic investigations will not require that corrections be applied to experimental data for the isotope effect of tritium because other factors associated with water flow are more significant.

Table 1.

Tritium and deuterium concentration ratios for Davidson clay and reference clay minerals. Deuterium measurements were not made for samples having missing data. The subscripts represent tritium (T) and deuterium (D) concentrations of tenaciously adsorbed water (aw), bulk pore water (bw), and water released at furnace temperatures of 110°C (adsorbed or bulk pore water), 300°C (after removal of 110°C water) and > 300°C hydroxyl water (collected after removal of 110°C and 300°C water).

	$\frac{T_{aw}}{T_{bw}}$	$\frac{D_{aw}}{D_{bw}}$	$\frac{T_{300}}{T_{110}}$	$\frac{D_{300}}{D_{110}}$	$\frac{T_{>300}}{T_{>110}}$	$\frac{D_{>300}}{D_{>110}}$
Illite	1.00	-	0.52	-	0.11	-
Montmorillonite	1.00	0.98	0.61	0.60	0.46	0.45
Kaolinite	1.00	-	-	-	0.02	-
Vermiculite	1.01	0.98	0.71	-	0.41	-
Halloysite	0.99	0.94	0.51	-	0.20	-
Davidson clay	0.98	-	0.39	-	0.14	-
Davidson clay (40°C equilibration)	1.01	0.99	0.48	0.42	0.21	0.20
Davidson clay (60°C equilibration)	1.06	1.01	1.05	0.45	0.47	0.21

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FORCED EXCHANGE OF TRITIATED WATER WITH CLAYS

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Isotopic fractionation is of fundamental importance, both for research techniques developed in recent years, e.g., tracer studies, paleoclimatology, origin of formation waters, and for technological developments, e.g., nuclear waste disposal, use of nuclear explosions for reservoir development and for civil engineering works.

Tritium Exchange and Fixation in Soils

When tritiated water comes in contact with earth materials, some of the tritium atoms may be retained preferentially (with respect to protium) on the minerals, especially on clays. Several mechanisms of retention are known to occur, or at least are theoretically possible. Some of these are: exchange of tritium ions for hydrogen ions in exchange positions, exchange of tritium ions for exchangeable cations other than hydrogen ions, tritiated water molecules exchanged for ordinary water of crystallization or hydroxyl water, and replacement of lattice aluminum sites by tritium (the aluminum ions moving into exchange positions). Depending on the process parameters (such as mineral species present, pH, ionic exchange capacity, isotope concentration, etc.), one or the other of these mechanisms may predominate. This exchange may cause a transient retardation of the isotope's flow through the soil. If the interaction of soil and isotope is very strong, a more or less permanent fixation of the sorbed isotope may take place.

Tritium has been called the single most hazardous byproduct of reactor operation [1]. As technological applications of nuclear energy increase, the production of tritium and its release into the environment are expected to increase sharply. Thus, the ability to predict quantitatively the effects of retardation and fixation of tritium in a given environment is crucial for planning of radioactive waste disposal. Yet little quantitative information is available concerning tritium exchange with and fixation by earth materials. As a result, such predictions are difficult if not impossible in most cases.

The fractionation of tritium by soil or reservoir minerals has been the subject of divergent opinions. Some investigators [2-5] have stated that the fractionation is minimal. At most, in the view of these investigators, clay minerals can produce a transient retardation of relatively minor significance [6]. For instance, Corey and Horton [4] and Corey and Fenimore [7] investigated the movement of tritiated and deuterated water through acidic kaolinitic soil and found no appreciable fractionation. These authors did not, however, monitor cation exchange or pH, nor did they analyze the clay itself for tritium content. The results of other investigators show, by contrast, that hydrogen isotopes may be selectively fixed [8-13]. However, neither are reaction rates well known nor are the directions fractionation may take when several isotopic species are present [14].

Reasons for the apparently contradictory findings are several. There is, first of all, the difficulty of modeling in the laboratory such an enormously complex system as is the soil and the flow of water through it. Secondly, certain important parameters, such as initial and final pH or the exchange of all major cations, were not monitored in some experimental studies. Thirdly, to the present writers' knowledge, no attempt was made in any previous work to establish a complete material balance for tritium by measuring the tritium activity in both water and the clay (or soil materials).

A further complication is the difficulty to distinguish between isotope exchange (a reversible process) and fractionation or fixation (essentially irreversible) in a short-term experiment in which both processes are going on simultaneously. The difficulty becomes particularly apparent when one considers the extremely small fraction of the total of available "exchange sites" that actually are part of these processes. This is clearly illustrated by the data presented in this study, see Table 4.

From the preceding discussion, based on a comprehensive literature search, several conclusions may be drawn:

a. Tritium may be fixed by earth materials under circumstances that are not well known at present.

b. It is at present not possible, even for idealized (simplified) cases, to predict quantitatively the isotopic fractionation (fixation) of the hydrogen isotopes under specified conditions of concentration, rock composition, pH, temperature, and pressure.

c. No data at all are available on the exchange rate of tritium and deuterium with reservoir materials.

Electrodialysis

Electrodialysis is a laboratory technique that has often been used to speed up ion exchange processes (hydrogenation) in clays. Its use has been criticized on the grounds that it leads to breakdown of the clay minerals themselves and hence "confounds" results. This objection is no doubt justified when extreme forms of electrodialysis are used. A careful examination of the facts suggests, however, that mild forms of electrodialysis are comparable to treatment of the clay with an acid of strength equivalent to the final pH reached in the electrodialysis. Since certain clays (notably the kaolins) are formed and are stable in a naturally acid environment, the objection to an appropriately mild form of electrodialysis appears to have no basis in fact. In the present writers' view, it is not likely that the weak electric field of the electrodialysis system (2 V/cm in the work discussed in this paper), as distinct from the electrodialysis-induced ion migration itself, can have a disruptive effect on the clay lattice. In support of this view, Marshall [15] points out that kaolinite experiences the smallest reduction in exchange capacity due to liberation of aluminum by salt treatment (following acid treatment). Kaolinite is chemically the most resistant of common clays in acid and alkaline solutions, and for certain clays electrodialysis is a less drastic treatment than leaching with acids of moderate strength. This appears to contradict the findings of Low [16] and Coleman and McAuliffe [17]. Low found that electrodialysis had a higher attack efficiency than acid treatment.

However, his potential gradient was about twice that used in the present study and, perhaps even more decisive, his final pH was much lower (2.7 to 3.0 vs. 4.5 to 5.0). Coleman and McAuliffe present data on electrodialysis that indicate a release of aluminum from two bentonite samples, similar in magnitude to the release achieved by treatment with 0.05 N HCl (equivalent to a pH of about 1.3). Coleman and McAuliffe do not indicate the potential gradient, length of electrodialysis, and final pH reached in their electrodialysis tests, so that a comparison with other results is not possible.

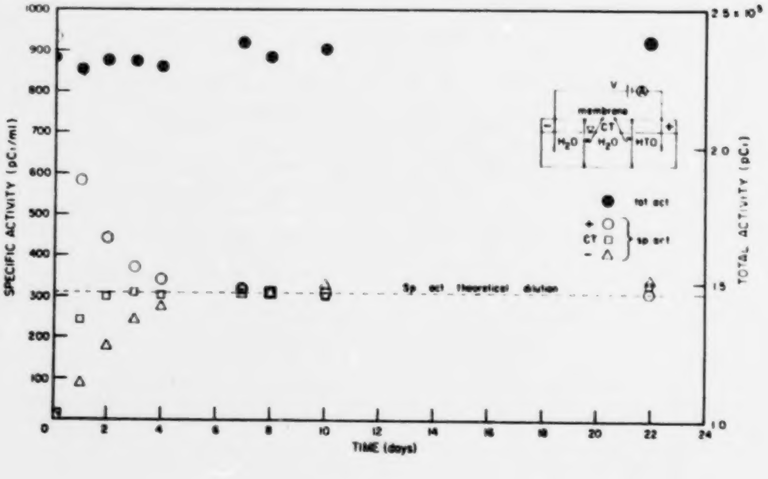
Procedure

Reference clay minerals were used in this study [18]. Kaolinite from Macon, GA (American Petroleum Institute No. 4), kaolinite from Mesa Alta, NM (API No. 9), and illite from Fithian, IL (API No. 35), were obtained from Ward's Natural Science Establishment, Rochester, NY. The clay samples were oven dried at 100°C, ground, and sieved between 53 and 74 microns, with no additional treatment. One hundred g portions of the clays were used in all of the runs.

The clays were electrodialyzed up to 54 days. The three-compartment electrodialysis cell [19] is shown in Figure 1, inset. All

Figure 1

Electrodialysis Without Clay. Tritium Activities as a Function of Time. Standard Deviation ±4%



experimental runs were conducted under the same conditions. The center compartment of the electrodialysis cell was loaded with the clay sample and 200 ml of ^1H_2O. The anode and cathode compartments were filled with 250 ml of tritiated water (917 pCi/ml;8 X 10^{-9} meq of tritium) and ^1H_2O, respectively. The clay was saturated and just covered with water. The voltage gradient was kept at 2 V/cm. Deionized distilled water was used for the dilution and background measurements.

During the course of the experiments, the fluid from each compartment was analyzed periodically both for tritium and ionic composition. The pH of each compartment was measured once every 24 hours after stirring the contents. At the completion of each run, the liquid in each compartment was analyzed for Na^+, K^+, Mg^{++}, and Ca^{++}.

Tritium was recovered from the clays by heat treatment followed, in some cases, by digesting with acid. The acid treatment was used because the tritium recovered by heating the clays to 670°C was much less than the tritium lost from the water in the compartments. First the clay-water mixture was transfered into a vacuum distillation apparatus where adsorbed water was removed from the clay at 110°C. The distillation was then repeated in a vacuum-sealed oven in steps of about 100°C up to 670°C. The temperature was held constant for three hours at each temperature. The evolved water was collected in a liquid nitrogen trap. After extraction, the tritium concentration of the recovered lattice water was measured. For the acid treatment, sealed containers with 1.0 g of clay samples (before and after electrodialysis) and 4.0 ml of $12\underline{M}$ HCl were heated to 200°C for 12 hours. This process assured the decomposition of the clay and released fixed tritium. The mixture was then vacuum-distilled and the tritium concentration of the distillate was measured.

Portions of the clays were leached with $1\underline{N}$ $BaCl_2$ (before and after electrodialysis) in order to determine the amount of exchangeable Al^{+3} in the clay.

Measurements

The tritium activity of the water in the three cell compartments, was measured either with a low-level internal gas counter or a liquid scintillation counter (Packard, Tri-Carb, Model 2111). The clay extracts were analyzed by gas counting techniques only. One-milliliter samples of liquid water are required with either counter. The background count for the gas and liquid scintillation counters was 2 cpm and 26 cpm, respectively. For one sigma confidence level, the error in the specific activities was ±2% for the gas counter and ±4% for the liquid scintillation counter. Systematic measurement errors were minimized by counting samples at random and not in the time sequence in which they were taken (and in which they are plotted). Therefore, the confidence levels of ±2% and ±4% are statistically valid.

Cations in the water samples were measured with an Atomic Absorption Spectrophotometer (Perkin-Elmer 303). From this, the total cation-exchange capacity (CEC), in milliequivalents, was calculated. X-ray diffraction measurements (Norelco, X-ray Diffractometer) of the clays also were done before and after each run in order to determine any alteration in lattice spacing under electrodialysis.

Differential thermal analysis (DTA) was used to determine the effectiveness of the heat treatment in recovering the hydroxyl water. In addition, clay samples were checked by this method after the acid treatment. Detail of the experiments and analysis techniques are described by Rabinowitz [20].

Results

The results are presented in Figures 1 through 5. Although all runs were conducted under similar conditions, there are essential distinctions between the experiments that affect the analysis of the results. The discussion of each individual figure will develop the pertinent differences between the experiments.

Control run

Figure 1 shows an electrodialysis run made without clay in the center compartment. This run was conducted to ascertain the accuracy of the counting procedure used in all experiments and to determine any possible tritium losses. The three curves represented by open symbols show the changes in tritium concentration or specific activity for the three cell compartments as a function of time. The specific activity at theoretical dilution, indicated by the dashed horizonatal line, was calculated from the initial total volume of water in the cell and the initial specific activity of tritium. Total activity, at any time, is represented by the solid symbols and is given by

$$T = \sum_{i=1}^{3} C_i V_i \qquad (1)$$

where T is the total activity (pCi) at time t, C_i is the tritium concentration (pCi/ml) in compartment i, and V_i is the volume of water (ml) in the same compartment. The total activity curves are corrected for the activity removed from the cell by sampling and by evaporation. Evaporation did in no case exceed one ml/cell-day. Corrections for electrodialysis are negligible.

Figure 1 clearly demonstrates that no tritium was lost from the water to the cell materials under the electric field. After eight hours the current in the cell remained constant at 3 ma. The pH, after the first day, stabilized at 2.9, 3.4, 10.0 in the anode, center, and cathode compartments, respectively.

Equalization in tritium concentration across the cell was reached after six days. The slight increase in tritium concentration toward the end of the run is within experimental error.

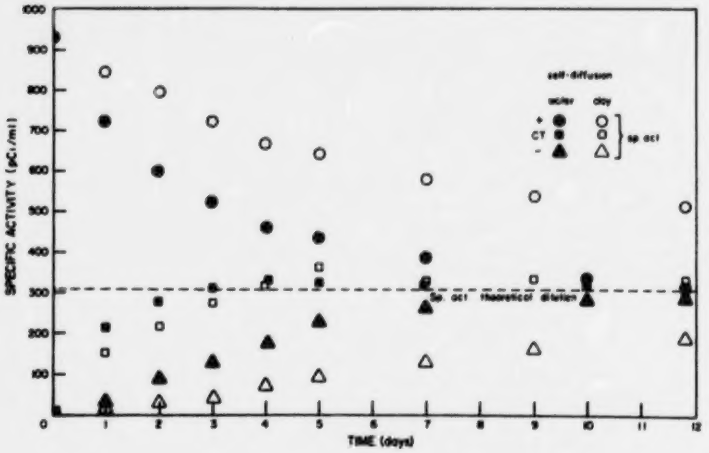
Self-diffusion

Figure 2 illustrates the results of two experiments conducted under self-diffusion of the tritium through the cell (no applied electric field). One run was made with water only (solid symbols). For the other, the center compartment was filled with a clay-water mixture (kaolinite No. 4). Only the first 12 days are shown in the figure. These data provide a comparison with the results obtained under electrodialysis. The pH of the individual compartments remained constant in both experiments. A representative tritium concentration of the clay in the center compartment (open symbols) was measured after the clay-water mixture was well stirred and allowed to settle back. This procedure was necessary because the tritium concentration in the clay was much higher than in the water immediately above the clay as long as equalization of tritium concentration across the cell had not been reached.

The retardation of tritium through the clay sample is very conspicuous. Equalization in the clay experiment was not reached until the 25th day. The specific activity curves for the cell containing only water indicate that the cell membranes have a negligible retarding effect on the trend toward equalization throughout the cell. No change in the total activity had been observed after 90 days when the two runs were terminated.

Figure 2

Self-diffusion Through Water and Through Kaolinite, API No. 4. Specific Activities of Tritium as a Function of Time. Standard Deviation ±4%

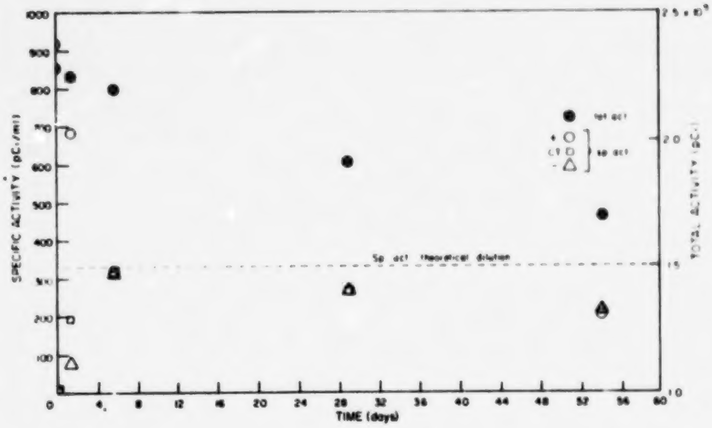


Kaolinite No. 4

Figure 3 represents the forced change in specific and total activity of tritium in the water during electrodialysis. Equalization in specific activity for the three compartments occurred just below the theoretical dilution value after only five days, or five times as fast as in the preceding case without electric field. The pH of the clay-water mixtures in the center compartment was 5.0 at the time of concentration equalization and dropped to 4.8 during the remainder of the run. Once the equalization of tritium concentration was reached, tritium began to disappear from the compartments at a constant rate of 60 pCi/h.

Figure 3

Electrodialysis of Kaolinite, API No. 4. Tritium Activities as a Function of Time. Standard Deviation ±2%

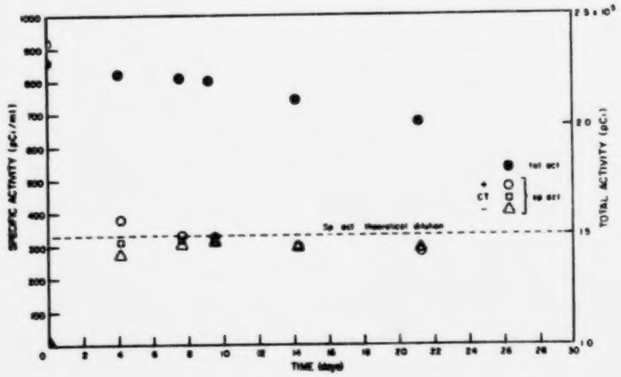


Kaolinite No. 9

Figure 4 represents the forced change in specific and total activity of tritium in the water during electrodialysis. In this run, equalization of specific activity was reached after seven days and at the theoretical dilution value. At that time the pH value in the center compartment was 7.5. The tritium disappearance from the water

Figure 4

Electrodialysis of Kaolinite, API No. 9. Tritium Activities as a Function of Time. Standard Deviation ±2%



did not begin until 60 hours after equalization had been reached, at which time the pH in the center compartment was 5.3, see Table 3. The rate of tritium loss in this run was 100 pCi/h.

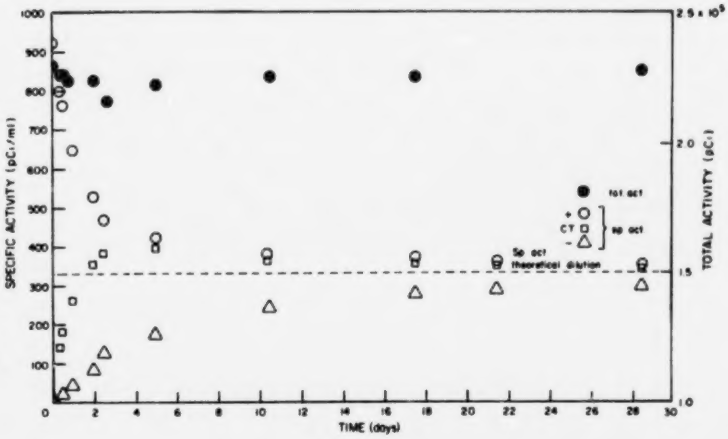
Illite No. 35

Figure 5 represents the forced change in specific activity of tritium in the water during electrodialysis. Equalization in specific activity between the anode and center compartments was reached after five days and was appreciably higher than the theoretical dilution value. Equalization across the cell was not reached even after 28 days, when the experiment was terminated.

The trend of the specific activity in the center compartment shows a strong retarding effect by the clay on the tritium migration under the electric field. In the previous experiments, the trend of the specific activity curves paralleled the trend in the total activity. In this experiment the total activity plot shows a correlation with a change in pH at about three days. There is a drop in total activity of 13,000 pCi and a corresponding change of pH in the center compartment to 3.6. Total loss after 28 days was well within the experimental error, however, see Table 3.

Figure 5

Electrodialysis of Illite, API No. 35. Tritium Activities as a Function of Time. Standard Deviation ±2%



Tritium concentrations in clay water

Tables 1 and 2 are a summary of the tritium concentrations and total activities measured in the interlayer and hydroxyl waters released from the clays at the various temperatures and in the acid distillate. The tritium concentrations for the three long runs (Figures 3, 4, and 5) were measured as composite samples over the interval 350°C to 670°C. The additional runs shown in Table 1 were conducted in order to confirm that the change in tritium concentration of the hydroxyl water is a time-dependent process even under electrodialysis conditions.

Ionic Composition

Table 3 is a summary of measured parameters. The recovered cations are the total of Na^+, K^+, Mg^{++}, and Ca^{++}. The total cations recovered and the specific conductance of the liquid in the center compartment were used as indicators of the completion of the electrodialysis process.

Exchangeable Al^{+3} was found in the kaolinite clays only. This amounted to about 20% of the exchange capacity of each clay.

Discussion

Tritium activity changes in the water

Even a casual comparison of Figures 3, 4, and 5 reveals that the loss in total activity with time differs strikingly for the three reference clays which were electrodialyzed under similar conditions. The total loss varies from almost 26% of the initial total activity for the Georgia kaolinite to less than the error of determination for the illite. In all cases, the total tritium recovered from hydroxyl water was less than 600 pCi (Table 2) and accounts for only a small fraction of the tritium lost from the water. The best fit for the total activity data in the control run is a constant within experimental error. The best fit for the total activity data for both kaolinite clays, after equalization in concentrations, Figures 3 and 4, is a straight line showing a constant rate of loss of tritium from the water to the clay.

The specific activity in both kaolinite samples approached the equalized values rapidly under electrodialysis. The specific activity was equal in all compartments after five days for the Georgia clay, Figure 3, and after seven days for the New Mexico clay, Figure 4.

For the Georgia kaolinite, the equalization value was lower than the theoretical dilution value, Figure 3. This suggests that exchange was proceding at a slow rate. After equalization had been reached, however, the exchange rate accelerated, as is evident from the slope of the specific activity vs. time curve.

An apparent preferential affinity of the clay for the tritiated water molecule prior to equalization in concentration was found when the water, in the center compartment, was first stirred above the clay and then stirred with the clay. The water sample taken above the clay was consistently lower in activity than the sample of water taken after mixing and settling. This effect was especially marked in the illite, Figure 5. The retarding effect of the clay accounts for the (small) decrease in total tritium activity in the water at the beginning beginning of the electrodialysis for each clay. This initial drop did not occur in the control run without clay.

pH and water conductivity of the clay-water mixture

The pH and the conductivity of the water in the center compartment, and the presence of exchangeable Al^{+3}, are the only parameters which show strong correlation with the tritium loss from the water, Table 3. The pH of the clay-water mixture in the center compartment for the Georgia clay was 5 at equalization of specific activities and remained 4.8 for the remainder of the run.

The pH of the water for the New Mexico clay did not become acidic until after nine days when it was 5.3. Loss in total activity was small until this time and then increased to 100 pCi/h.

In the illite run, the only apparent loss in tritium, after the initial drop in total activity, occurred after three days in coincidence with a change in pH from 6.4 to 3.6. After five days the pH in the center compartment had returned to 6.6, and the total tritium activity kept rising to the end of the run. No exchangeable Al^{+3} was found in this clay after electrodialysis.

No evidence was found that the pH change in the center compartment was related to any physical changes in the system other than the exchange property of the clay. Low [16] has discussed this phenomenon and drew the same conclusion. X-ray diffraction measurements of the reference clays before and after electrodialysis indicated no shift in lattice spacing or alteration of the basic structure of the clay minerals.

The water conductivity in the center compartment of both kaolinites approached low values compared to the end compartments, as equalization in tritium concentration was reached across the cell. Until equalization in tritium across the three compartments was reached, tritium fractionation probably was not appreciable. This is based on the fact that T/H approached the theoretical dilution value. After the major fraction of the cation exchange was complete, both the pH and the conductivity of the center compartment decreased. At this time, tritium began to disappear from the water in the clay compartment. This loss of tritium continued at a constant rate as long as an electric field was applied across the clay.

The illite behaved quite differently. Tritium equalization across the three compartments had not been reached after 28 days (termination of run); neither did the pH or the conductivity drop during that time.

Table 1

Tritium Concentrations in Water Released From the Clays as a Function of Temperature

Sample Number	Tritium Content in dis/min·ml X 10^4		Deviation, Gas Proportional Counting From NBS Value
	NBS Value	Gas Proportional Counting	
1	2.91	2.96	+1.7%
2	2.37	2.36	-0.4%
3	1.92	1.76*	-8.1%
4	2.26	2.27	+0.4%
5	2.79	2.74	-1.8%

*Low proportional count due to adsorption. See text.

Table 2

Tritium Activity Recovered by HCl Treatment After 12 h at 200°C with Clay Samples

Samples	Electrodialysis duration in days	Conc. recovered (nCi/ml/g)	Total act. recovered by heat treatment (nCi/100 g)	Net total act. recovered by HCl treatment (nCi/100 g)	Remarks
Kaolinite API No. 4	Fresh clay	Background	-	-	oven dried at 110°C
Kaolinite API No. 4	56 (Fig. 3)	6.6 (*)	470	2090	
Kaolinite API No. 4	56 (Fig. 3)	6.5 (*)	470	2130	preheated to 670°C before acid treat.
Kaolinite API No. 9	21 (Fig. 4)	3.6	160	1290	
Illite API No. 35	28 (Fig. 5)	2.0 (*)	590	210	

*Counted twice

Table 3

Summary of Measured Parameters

Sample	Cations recovered (meq/100 g)					Al^{+3}	Final pH			Final Conductivity (µmhos/cm)		
	Na^+	K^+	Mg^{++}	Ca^{++}	Total	(meq/100 g)	-	CT	+	-	CT	+
Control (tap water)	--	--	--	--	--	--	10.0	3.4	2.9	860	53	1150
Kaolinite API No. 4	0.2	1.1	1.0	8.2	10.5	1.90	10.9	4.8	2.1	795	156	3976
Kaolinite API No. 9	0.6	0.0	0.4	1.2	2.2	0.32	8.3	5.6	3.3	278	102	363
Illite API No. 35	1.8	2.4	1.2	29.2	34.6	0.01	10.6	6.6	2.3	3114	795	3892

Table 4

Analysis of Clays. Fractional Increase of Tritium Atoms in Al Positions Over Equilibrium Value

Sample	Number of exchange positions vacated by exchangeable Al per 100 g of clay	Net tritium recovered by acid treatment (T atoms) per 100 g of clay	Al positions filled by T atoms (%)	$\frac{(T/H)_{Al}}{(T/H)_{equilibrium}}$
Kaolinite No. 4 (Fig. 3)	1.1×10^{21}	4.3×10^{10}	4.0×10^{9}	400
Kaolinite No. 9 (Fig. 4)	0.2×10^{21}	2.7×10^{10}	1.4×10^{-8}	1400
Illite No. 35 (Fig. 5)	none	$< 3 \times 10^{9}$	none	no fractionation

Tritium recovered from the clay extracts

From Tables 1 and 2 it is clear that a considerable amount of additional tritium was released by digesting with acid a portion of kaolinite No. 4, Figure 3, that had first been heated to 670°C. DTA before and after each of the above treatments revealed: (1) holding the temperature constant for three hours at each temperature step was not sufficient to remove all of the hydroxyl water; (2) the exothermic peak at 950°C, characteristic for kaolinites, was flattened after the acid treatment.

Tritium activity in the hydroxyl water

Tritium exchange with hydroxyl groups clearly took place, as shown in Table 1. Furthermore, since the tritium concentrations of the recovered water decreased with increasing temperature, we can conclude that there are hydroxyl groups of different binding energy. Specifically, fractionaction was noted in the 110 to 150°C interval. The most striking result is the fact that hydroxyl exchange was proportional to cation exchange capacity of the three clays, to wit: Illite, 35 meq/100 g and 93 pCi/ml; kaolinite No. 4, 10 meq/100 g and 36 pCi/ml; kaolinite No. 9, 2 meq/100 g and 12 pCi/ml. Although some of the tritium concentrations in the hydroxyl water are high, the total activity recovered by heat extraction was less than 600 pCi for any of the clay samples.

Tritium activity recovered by HCl treatment

The release of tritium from the clay by digesting with HCl at 200°C indicates the existence of an exchange mechanism which is completely different from the hydroxyl exchange. Although the DTA data show that heating a clay to 670°C does not release all of the hydroxyl groups, there is evidence that such a large quantity of tritium, as was released by the acid treatment, could not have been bound in the 900 to 1150°C fraction [9].

The net total activity of tritium recovered (Table 2) could have been bound in one of two exchange positions in the clay: (1) cation exchange positions; (2) lattice (octahedral) positions vacated by aluminum which, under the electric field, had moved into exchange positions. There are indications [16] that H-Al exchange takes place in a clay under electrodialysis. The findings of our acid treatment together with the observed changes in tritium activity during the electrodialysis, and the exchangeable aluminum tend to support the second alternative.

The amount of tritium recovered by acid treatment from an unheated portion of the illite was 800 pCi as compared with 590 pCi recovered by heat treatment. The net gain by acid decomposition was, therefore, 210 pCi. No exchangeable aluminum was found by leaching the illite after electrodialysis. Even though there may have been some substitution of tritium for potassium, this substitution was in the same ratio of T/H as in the water and was not detectable in the total activity determinations. In clay No. 4, Figure 3, where 1.9 meq/100 g of exchangeable aluminum was found, the total tritium atoms present in the electrodialysis cell represent only 4 X 10^{-7}% of the total number of exchange sites available. This ratio for clay No. 9, Figure 4, was 2.4 X 10^{-6}%. In view of the large number of available sites, the energy required for fractionation under forced exchange would probably not be large.

The fractional increase in tritium atoms exchanged over equilibrium (eq) value is given by $(T/H)_{Al}:(T/H)_{eq}$, where (2) $(T/H)_{Al}$ is the ratio of T-atoms to H-atoms occupying the exchangeable Al positions in the clay lattice at the end of the process, $(T/H)_{eq}$ is the equalization value for T/H in the electrodialysis cell ($\approx 10^{-13}$ for all runs). Table 4 presents the pertinent numerical values needed to calculate Eq. (2) for the three long runs. From the change in total activities, Figures 3 and 4, the tritium exchange appears to be a linear process in time.

ACKNOWLEDGMENTS

Acknowledgment and appreciation is made to Professor Kay Brower who suggested and carried out the acid treatment of the clay. This work was supported in part by the New Mexico Water Resources Research Institute, Project No. 3109-114.

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DYNAMIC MODEL OF WATER MOVEMENT IN SOIL UNDER VARIOUS CLIMATOLOGICAL CONDITIONS

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The purpose of this study was to ascertain if a previously developed kinetic equation ecosystem model was sufficiently general to simulate and predict the movement of water (both stable and tritiated) in soil under diverse climatological conditions and, if so, to apply this model to water movement in the soil of a tropical rain forest, an Illinois grassland and a Nevada desert site. The model, called MESA

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* Work performed while on assignment with the Radiological Physics Division, Argonne National Laboratory.

(multivariable ecosystem analysis), has been presented and discussed previously [1,2]. The water in the soil is described in the model by 50 simultaneous first order, linear differential equations with discontinuous, variable coefficients. The coefficients are assumed to be functions of the evapotranspiration, soil-water diffusion, gravity flow of soil water, and the previous 48 hours history of rainfall.

Soil water had been found in earlier studies [1-3] to move downward in a pulse due to gravity flow (after a rainfall), to spread out due to diffusion, and to leave the system by evapotranspiration. It was an additional purpose of the authors to determine the significance of these parameters as a function of climatic variables.

Method

In order to provide some validation of the prediction model it was desirable that the study be made of sites where a) tritiated water movement studies have been made, b) where the required climatological data were available and c) where the climatology differs significantly. Three sites were found that fulfilled all the above requirements. They were: the Puerto Rico rain forest, the Illinois grassland and the Nevada desert.

The studies of tritiated water movement in the Puerto Rico rain forest have been made by Jordan, et al. [3], and the climatological data for these studies were obtained from El Verde Rain Forest Project [4]. The movement of tritiated water in the soil of Illinois grassland and the related climatological data have been reported previously [1,2].

The tritium inventory at site 18 A [5] of the Sedan ejecta five years after the detonation along with the pertinent climatological data were obtained from Koranda and Martin [6].

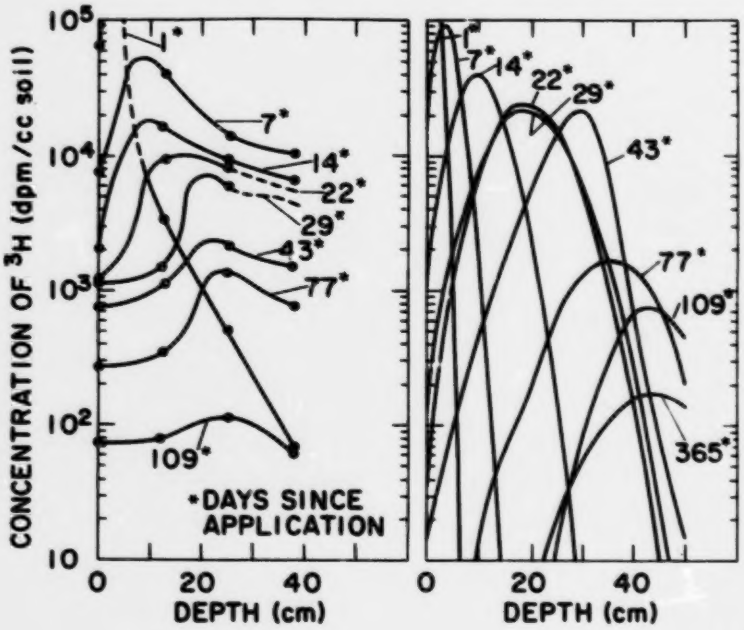
Results and Discussion

The tritium concentration in the soil of the Puerto Rico rain forest [3] is present in Figure 1a and the concentration as predicted by MESA in Figure 1b. One notes that basic agreement is obtained as to the location of the bulk of the tritium between the observed data and the prediction. For example, the peak concentration after 76 days is predicted to be at 28 cm in depth and to have a value of 5×10^3 dpm/ml. This compares well with an observed peak of between 25 cm and 30 cm and a concentration of 10^4 dpm/ml.

Tiitium concentration predictions were made until at least 99.9% of the tritium had passed through the Puerto Rico and Illinois ecosystems and at least 99% through the desert ecosystem. Using these criteria, the Puerto Rico rain forest predictions were made for one-half of a year, the Illinois grassland for one year and the Nevada desert for five years.

Figure 1

Concentration of Tritium in Puerto Rico Rain Forest Soil as a Function of Soil Depth. Figure 1a (left side) observed, Figure 1b (right side) predicted



The tritium concentrations, experimental and predicted, for Illinois grassland are given in Figures 2a and 2b respectively, and show good agreement between the two sets of curves as observed, at least until 77 days after application, at which time less than 0.17% of the tritium remained in the ecosystem. The data points showing tritium concentration five years after application to the Nevada desert are presented in Figure 3 with prediction curves, and show excellent agreement between the data points and predicted concentration.

The downward movement of the peak or maximum concentration as a function of time, presented in Figure 4, indicates that the downward movement of the peak is due primarily to the rain-induced gravity flow of the soil water. The peak essentially stops its downward movement when the bulk of the tritium is below the level of the soil where rain induces a moisture concentration in excess of the field capacity. It can be seen that in the Puerto Rico rain forest, where the rain exceeds 100 inches per year, the peak starts movement at a very high rate (up to 2 cm/day) but after 45 days, when the peak is approximately 30 cm in depth, the rate is 0.2 cm/day or less. This large change in the rate of peak movement was observed and reported by Jordan et al. [3].

Figure 2

Concentration of Tritium in Illinois Grassland Soil as a Function of Soil Depth. Figure 2a (left side) observed, Figure 2b (right side) predicted

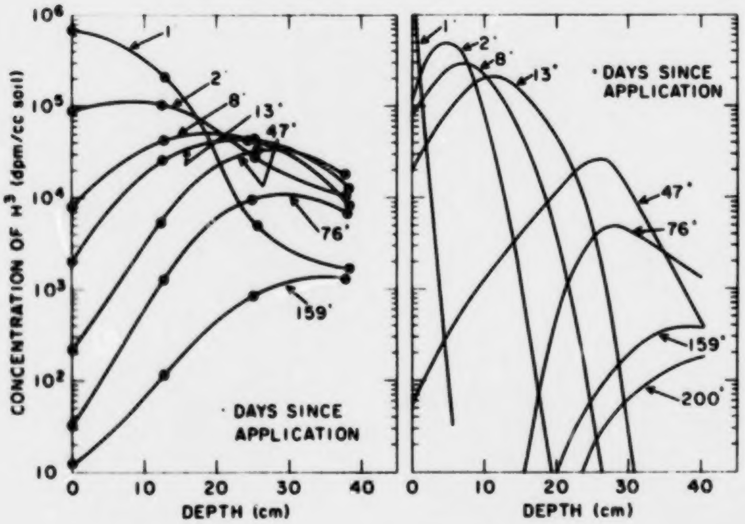


Figure 3

Predicted and Observed Concentration of Tritium in Nevada Desert Soil as a Function of Soil Depth

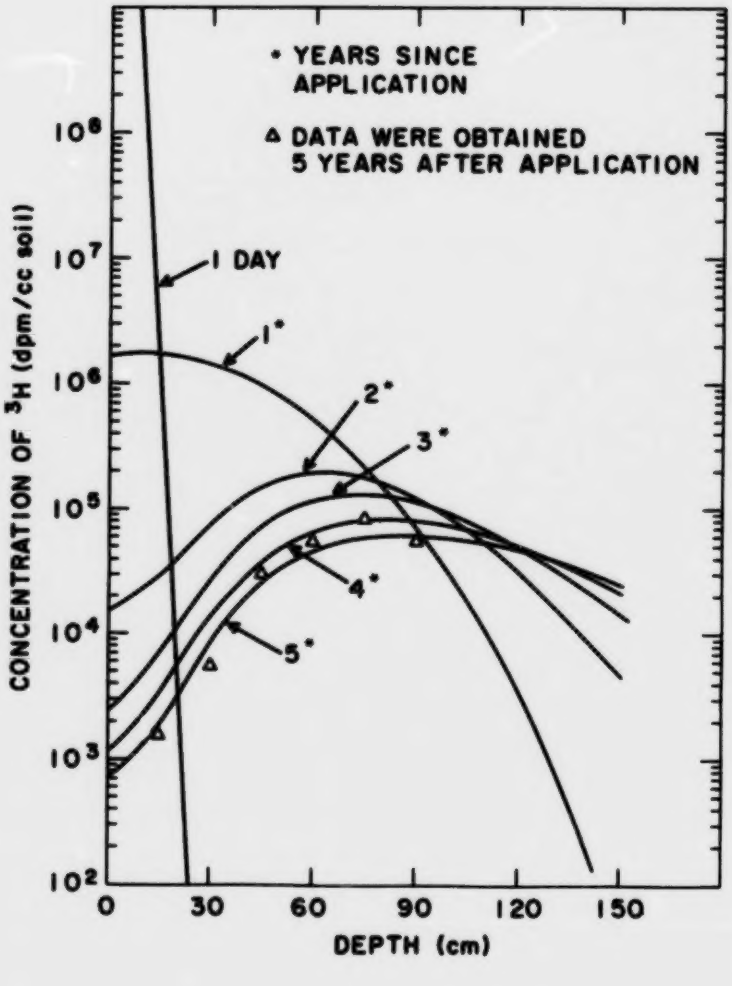
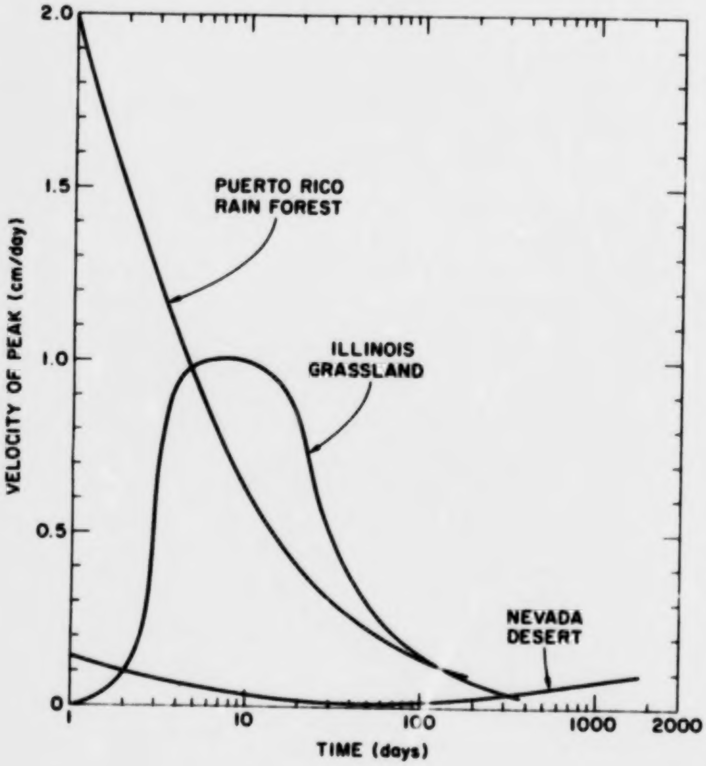


Figure 4

Downward Velocity of Peak Concentration as a Function of Time After Application



They also observed a soil relatively impervious to water below approximately 30 cm in depth. Thus, the downward movement of the peak stopped only when the peak reached a soil that was impervious to water.

In the Illinois grassland curve, Figure 4, the "hump" occurs during the spring when the soil is saturated. Very little movement of the bulk of the tritium occurs at other times.

For the Nevada desert, where rain is rare, the downward bulk movement of tritium is exceedingly slow and probably is not due to rain-induced gravity flow. Even if there is no rain, the bulk of tritium will eventually be below the plant roots. This can be explained as follows: a) diffusion will move some of the tritum deep into the soil and b) transpiration will move tritium out of the upper part of the soil. Thus, the bulk of the tritium will eventually be below the level of the plant roots even if no gravity flow occurs.

The total amount of tritium in the soil, i.e., integrated over all depths, as a function of time is shown for the Puerto Rico rain

Figure 5

Total Tritium per cm^2 of Surface in the Puerto Rico Rain Forest as a Function of Time After Application

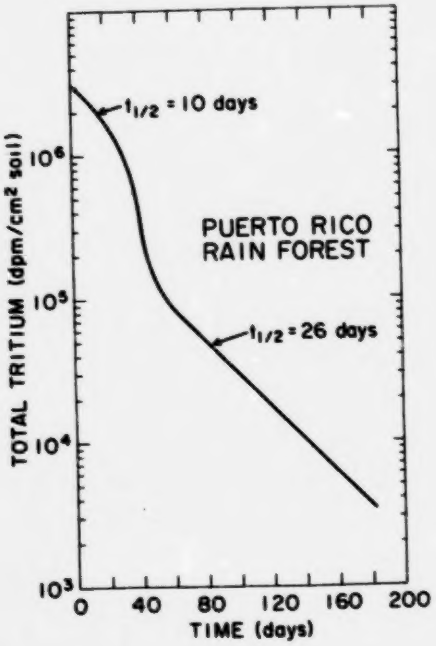


Figure 6

Total Tritium per cm^2 of Surface in the Illinois Grassland as a Function of Time After Application

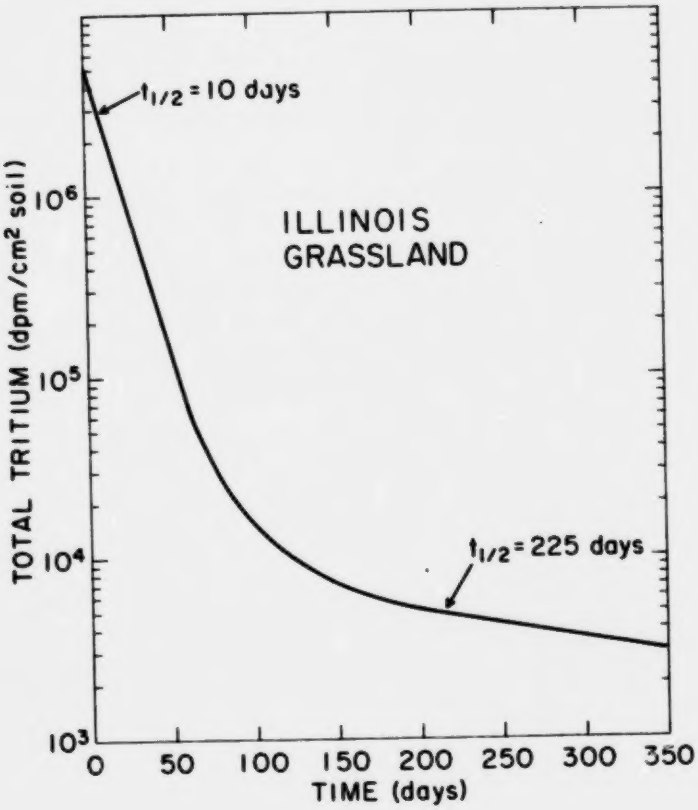
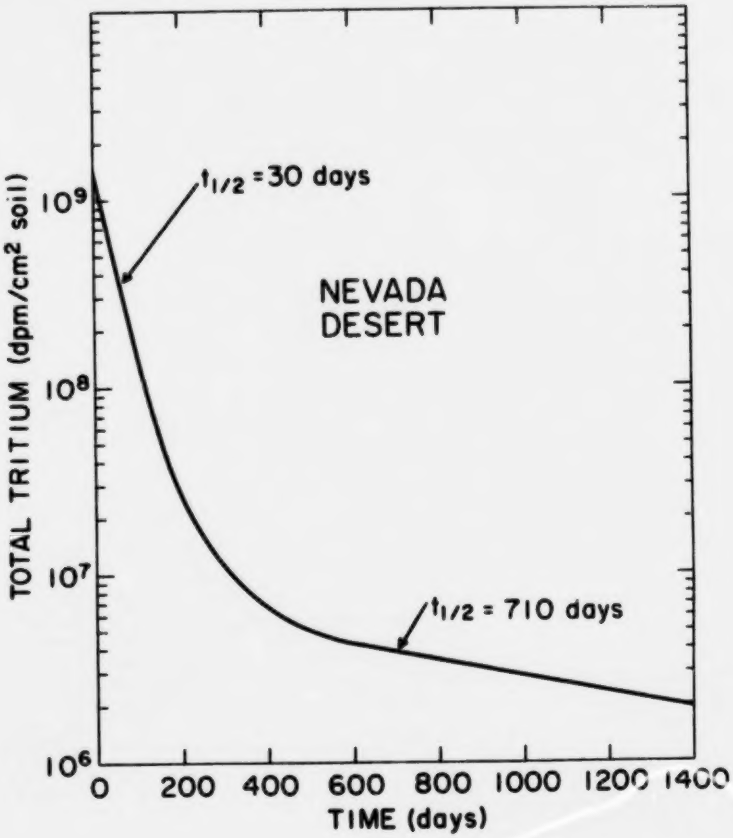


Figure 7

Total Tritium per cm^2 of Surface in the Nevada Desert as a Function of Time After Application



forest in Figure 5, the Illinois grassland in Figure 6, and the Nevada desert in Figure 7. The significant aspects of these curves is that they predict not one, but two distinct half-residence-times for tritium in soil. The initial value of the half-residence-time occurs when the bulk of the tritium is in the region of the plant roots where tritium is removed from the soil by transpiration of the plants. Once the bulk of the tritium is below the plant roots (50 days for the rain forest, 150 days for the grassland and two years for the desert) the half-residence-time increases by about an order of magnitude, except for Puerto Rico where the lateral drainage keeps the half-residence-time low. This phenomenon is explained by noting that, whereas transpiration removes tritium from the upper soil, tritium is moved from the lower to the upper soil by diffusion. That is, the program predicts that as long as the bulk of tritium is in the region of the plant roots, the rate of transpiration will be the controlling factor in determining the half-residence-time of the ecosystem. However, once the bulk of the tritium is below the root area, diffusion, a much slower process than transpiration, will become the principal factor in determining the half-residence-time.

Martin, et al. [7] report half-residence-time in the Puerto Rico rain forest of 29 ± 1 days, a value in substantial agreement with the 10 to 26 days predicted in this study. For the desert, a half-residence-time of 400 ± 40 days was predicted [5]. This differs considerably from our predicted value of 30 days for the first year and 700 days after two years.

Conclusions

1. In the Puerto Rico rain forest, the most significant parameter controlling water (stable and tritiated) movement through the ecosystem is the rainfall.
2. In the Illinois grasslands, the rainfall and transpiration rate are the primary factors controlling water movement through the ecosystem.
3. In the Nevada desert, the half-residence-time is primarily influenced by the transpiration and diffusion rates.
4. In predicting long-term concentrations of tritium in the soil (other than possibly a tropical rain forest soil), it may prove an advantage to consider the tritium movement in the ecosystem as characterized by two separate half-residence-times. The initial value is determined primarily by the rate of transpiration and the final value, about an order of magnitude greater, is determined by the soil-water diffusion rate.
5. The ecosystem model, MESA, appears to be sufficiently general to give satisfactory predictions of water movement in a tropical rain forest, grasslands and desert soil.

ACKNOWLEDGMENT

This work was performed under the auspices of the U. S. Atomic Energy Commission.

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CHAPTER SEVEN
Environmental Monitoring Techniques

MONITORING REACTOR EFFLUENTS FOR TRITIUM: PROBLEMS AND POSSIBILITIES

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There are two reasons for monitoring effluents for the presence of tritium. The first is to be able to measure the total release of tritium to the environment, thereby ensuring that chronic releases are at acceptable levels. What is "acceptable" may be defined by health considerations, e.g., the permitted release of tritium per se, or a more stringent requirement may arise indirectly, e.g., from the economics of heavy water loss in the case of heavy water cooled and/or moderated reactors. The second reason is to be able to measure the rate of release of tritium. The aim here is to be aware of any change in the rate of release above the normal base release. Such a change may indicate a mechanical failure, e.g., a cracked heat exchanger. The response times for monitoring systems measuring the rate of release will have to be shorter than for systems measuring the total release but, correspondingly, the concentrations of tritium of interest in the effluent may be higher.

The extremes of the practical compromise between response time and sensitivity are represented by these two monitoring purposes. Ideally one instrument should have the capability for covering this range for a given type of effluent but, as we shall see, although this may be possible it may not be economically justifiable.

General Requirements

The requirements for tritium monitoring methods for power reactor systems are far more stringent than those for laboratory or even research reactor installations [1,2]. In addition to meeting the appropriate sensitivity and response time requirements, the following specifications have to be met:

-Industrial quality rather than laboratory quality should be the rule. This means that the instrument must be mechanically robust and capable of operating in a wider range of temperatures and humidities than may be normal in the laboratory. A military type of specification may be appropriate.

-The instrument should have few controls and the function of these should be obvious to casual users such as power station operating staff (as opposed to instrument technicians, etc.).

-Continuous 24 hour/day operation should be possible with few or no manual adjustments.

-Calibration and baseline of any measuring system should be easy to establish in situ.

The requirements noted, so far, apply to any reactor or industrial-quality instrument. Another general requirement is that the response should be specific, in this case to tritium. The output should therefore be independent of: Nonuniform, time-variant gamma fields, certainly at the mR/h level; other radionuclides in the media being sampled; other nonradioactive contaminants such as dust particles, ions or luminescent materials; humidity, temperature, pressure; and previous measurements, i.e., the memory should be small compared with the range of variation expected in monitored activity.

The requirement for specificity of response has been, in practice, the hardest one to meet. Much of this paper will be concerned with efforts to meet this requirement. This work has been concerned entirely with tritium in the form of the oxide so that some of these remarks are not applicable to monitoring for elemental tritium.

Monitoring Water Effluents

Scintillation methods appear, at the moment, to be the only practicable approach to monitoring water effluents. Since the number of tritium betas reaching a solid/water interface is only 1.036/s cm^2 from a concentration of tritium in the water of 1 μCi/ml [3], liquid scintillators, with intimate mixing, possess an obvious advantage over solid scintillators.

Counting with Plastic Scintillator

To attain a high value for the product of counted volume and counting efficiency (defined here as counts per tritium beta entering the scintillator) small crystals of anthracene (presenting a high surface-to-volume ratio) have been used for many years. Detectors of this type have not been very successful for industrial application since they tend to be easily blocked by sedimentation and by filtration from the sample. Also, because of the tortuous light paths, they suffer rapid drops in counting efficiency with discoloration of the anthracene surface. Various kinds of detectors using thin sheets of plastic scintillator or anthracene spread over lucite rods have been described [4,5,6]. The properties of two detectors that have been developed are shown in Table 1.

Table 1

Properties of Plastic* Scintillator
Detectors for Tritium in Water

	I	II
Scintillator Area	3000 cm^2	240 cm^2
Size	5 X 5 X 5 cm cube	4.6 cm long 2.7 cm dia.
Response	24000 cpm per μCi/ml	39 cpm per μCi/ml**
Background Counting Rate	18 cpm***	8 cpm**

* Nuclear Enterprises NE102

** Detector in Nuclear Chicago Mark I liquid scintillation counter

*** Detector in 10 cm lead

Detector II has the same outside dimensions as a standard liquid scintillation counting vial and is shown in Figure 1. The counting efficiency, defined as above, is 26%. This design is being used in a commerically available, bench-type, liquid scintillation counter for effluent monitoring. In increasing the size of the detector (and scintillator area) to attain a better sensitivity (i.e., counting rate per unit concentration), the increased counted volume is traded off against lower counting efficiency. The larger detector, shown in Figure 2, represents such a compromise. The scintillator area is 3000 cm^2, the counting efficiency is 12%. In use as an effluent monitor, the detector is housed inside 10 cm of lead. The electronic analyzer is made from NIMs, the units being arranged as shown in Figure 3. The monitor has a most sensitive range of 0 to 10 nCi/ml and a response time constant of 35 s when the flow rate is 10 to 20 ml/s.

In operating with this type of detector in several nuclear power stations, a number of problems have had to be overcome. First, effluents are usually dirty. Filtration down to 5 or 10 μm pore size has been adequate to prevent crudding up the detector, though in one case dissolved iron complexes have discolored the scintillator over a period of weeks. Regular cleaning of the detector with mild complexing

Figure 1

Small plastic scintillation detector for tritium in water. The unit fits into a standard liquid scintillation counter.

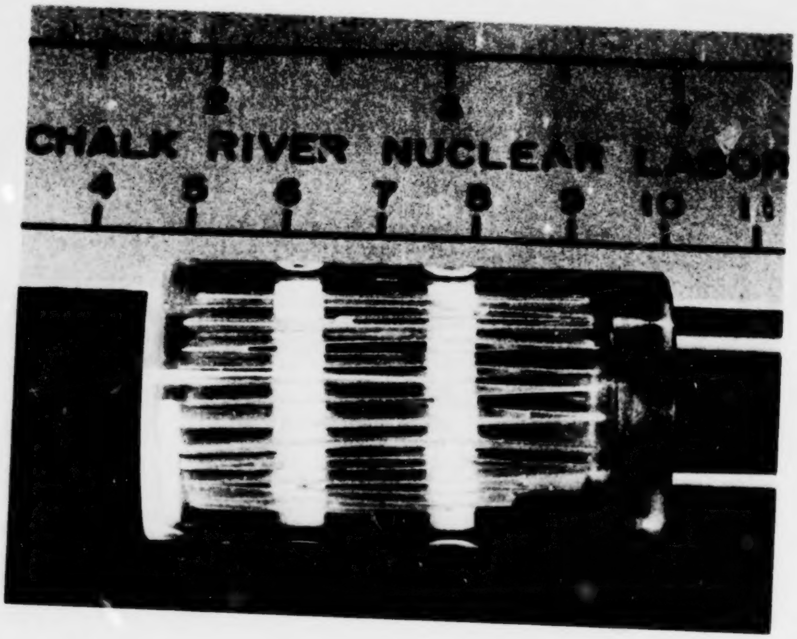
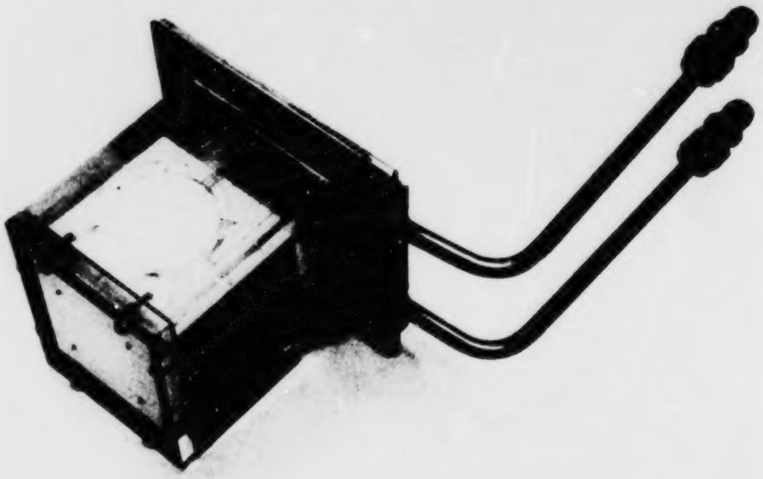


Figure 2

Large plastic scintillation detector for tritium

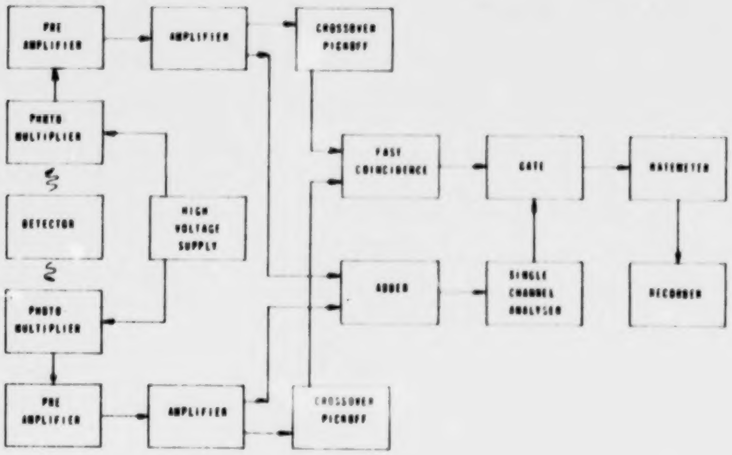


agents, such as citric acid, appears to be the answer to this. With some effluents, bacterial growth on the scintillator sheets has occurred but can be deterred by maintaining a sufficiently high flow rate through the detector. A preventive measure that has not as yet been tried is to expose a metallic silver surface to the water in the detector. Chlorination, which is normally used to prevent bacterial growth in water systems, interferes with the measurement of tritium through the chemiluminescence created. The photon emission rate is sufficiently high, even at low chlorination rates, for the random coincident counting rate between the two photomultipliers to be observable on the monitor output. Where the sampled system has been chlorinated (particularly in pulsed chlorination systems) a delay has to be introduced into the sample line or an activated charcoal filter has to be installed. The latter is preferable when a delay of approximately 20 minutes is not tolerable but, of course, it becomes another item needing regular maintenance.

Despite 10 cm of lead shielding, the monitor will respond to high energy gamma irradiation. For example, the response (in terms of equivalent tritium concentration) to ^{60}Co gammas (1.333 and 1.173 MeV) is 5 nCi/ml per mR h^{-1}. (At lower energies, for example 0.662 MeV, the response is $<$0.07 nCi/ml per mR h^{-1}.) The response to radionuclides in the sampled water is more serious. This can be reduced in two ways: Removal of the radionuclides before the water reaches the detector, and reduction of the efficiency of the detector for radionuclides other than tritium. Ionic products can be removed by ion exchange column, though this is expensive both in terms of material cost and servicing time. Gaseous products can be purged from the water by blowing air through the sample prior to passing it through the detector. The

Figure 3

Arrangement of nuclear instrument modules (NIMs) in tritium analyser. Bipolar pulses from the amplifiers go to the crossover pickoffs. Coincidences between the two channels within 20 nsec are determined in the fast coincidence unit and a gating signal applied to the gate for each coincidence. Unipolar pulses from the amplifiers are added and the sum pulse height analysed. The analyser output then passes through the linear gate, provided a gating pulse is present from the coincidence circuitry and drives a linear ratemeter.



response of the detector to beta/gamma-emitters in the water varies with the energy of the radiation. For example, the response is 3600 cpm per nCi/ml for ^{204}Tl beta energies, rising to 16000 cpm per nCi/ml for high (2.2 MeV) beta energies. The effective source volume for the other nuclides is very much higher than for tritium so that, although most of the scintillator pulses are larger than those from tritium and are rejected by the electronics, sufficient numbers fall into the tritium energy channel to give the responses noted above. With an extremely thin scintillator (comparable to the tritium beta maximum range) the situation would be worse. Reduction of the water volume in the detector is needed but is mechanically difficult to attain.

Automatic Counting with Liquid Scintillator

The minimum activity detectable by the plastic scintillation detector (1 nCi/ml) is not adequate for some purposes (for example, demonstrating conformity to the proposed guidelines for tritium in effluents from light water cooled reactors [7]). Counting with liquid scintillator of samples collected manually permits measurement of tritium down to pCi/ml and this method is several orders of magnitude less susceptible to interference from other nuclides, since the counting efficiencies and effective source volumes for tritium and the other nuclides are similar. Infrequent manual collection from an averaged or integrated sample may be adequate for the purpose cited but for response times less than a few hours, an automatic liquid system may be preferred.

Several years ago, this laboratory developed an automatic counter for determining the tritium content of urine or any aqueous sample with liquid scintillator [8,9]. Here, the instrument collects, on demand, part of the aqueous sample which it then mixes with liquid scintillator and counts. The instrument is also programmed to periodically measure standard and background solutions and can equally well be programmed to sample from any input at a frequency up to 20 to 30 times per hour. The reproducibility of the processing system is ±7% for water samples. The sensitivity was 120 cpm for 1 nCi/ml on a background counting rate of 60 cpm. The sensitivity could easily be improved by more than an order of magnitude by changing to a more sophisticated analyzer such as that in the plastic scintillation monitor or a commercial liquid scintillation counter. Important practical features in an instrument of this type, which are included in this monitor, are adequate containment of the new and used liquid scintillator, automatic detection of a fault in sample processing and an early warning of depletion of the scintillator or standard supply.

An alternative approach has been taken by Ting and Sullivan [10]. They have described a flow cell which will mix scintillator and sample supplied to it continuously and which, if attached to a suitable vacuum system, batches the mixture and transfers the batches into and out of a counting vial in a standard liquid scintillation counter. With one of their cells (Beckman Discrete Sampling Flow Cell), it was found in the laboratory that the reproducibility of this method was ±2.2% at a batching rate of 10 samples per hour. This reproducibility includes that of the sample and scintillator metering, which in this case was with a Technicon auto-analyzer multichannel pump. Tubing failure in the latter has so far limited the length of continuous (24 hour/day) unattended runs to 54 hours.

The sensitivity for the mixing ratio used (15:1) was 600 cpm for 1 nCi/ml on a background of 50 cpm. A suitable quality of metering/

containment system for this flow cell has not yet been engineered.

A serious disadvantage with a liquid scintillation-based monitor is the cost of liquid scintillator. A sample every 10 minutes costs about $6,000 per year in scintillator alone. If a longer response time is acceptable then sampling may be at a lower frequency (and costs correspondingly lower). However, the flow rates for continuous operation under these conditions are impracticably low (e.g., less than 0.3 mm^3/s for the sample) so some type of "on demand" processing, as is provided on the analyzer for urine, is more appropriate.

Monitoring Air Effluents

From the point of view of sensitivity to tritium, the choice of method or monitor type is wide [11,12]. Ionization chambers are capable of measuring down to 1 pCi/ml, proportional counters (such as the Berthold model [13]) to 0.5 pCi/ml, scintillation counters (such as detector I, Table 1, described for monitoring water) to 0.1 pCi/ml and a Geiger counter [14] down to 10 fCi/ml. Lower concentrations may be measured by collecting the tritium in the form of water and detecting it with liquid scintillator [15]. Gamma background and radionuclides in the sampled air pose a more severe limitation on the direct methods than the inherent sensitivities of the monitors to tritium.

In ionization chambers, the effect of gamma background is the lesser of the two evils. The interference (1 mR/h is equivalent to 0.1 nCi/ml) can be reduced to 1 to 2% by use of a sealed adjacent chamber [15]. The limitation here is not so much the mechanical problem of ensuring identical irradiation of the pair of chambers, as Jalbert and Hiebert [16] have recently demonstrated, but more that of maintaining identical air masses within the chambers despite variations in the pressure and temperature of the sampled air. In an installed effluent monitoring system, such as that being discussed here, sufficient lead shielding in conjunction with a sealed compensating chamber and sensible siting can eliminate gamma background as a limiting problem in most cases.

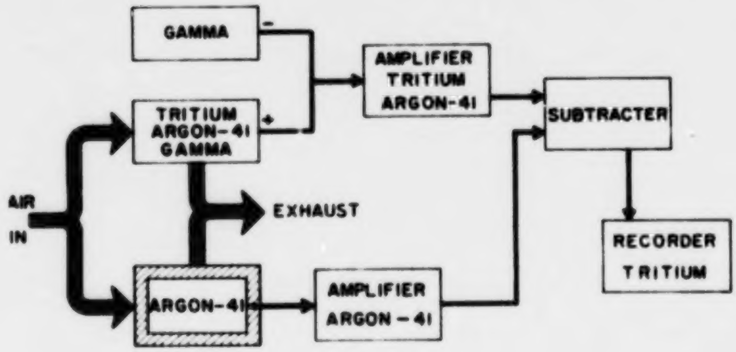
Interference from Other Radionuclides

Radionuclides in the monitored effluent have been the major problem. As in the case of water effluents, activation products such as ^{41}A, gaseous fission products (^{133}Xe, ^{85}Kr, etc.) or particulates may be present. The interference from the noble gases is the most difficult to eliminate; filtration (with activated charcoal for iodine if necessary) can eliminate particulates. Because of the greater energy deposited in a large ionization chamber per disintegration, the response of this type of monitor to the beta-emitting noble gases is greater than to the same concentration of tritium. For example, in a 40-liter ionization chamber, ^{41}A produces five times the ionization produced by tritium. The relative responses are dependent upon both chamber geometry and beta energy: With smaller chambers the responses are more nearly equal.

By measuring the ^{41}A activity alone, a signal can be obtained for subtraction from the gross output. For example, Soudain [17] used a Geiger counter in the side of an ionization chamber. In this study a scintillation counter sampling the air in parallel with a gamma-compensated ionization chamber has been used, see Figure 4. However, because of the different relative responses to different beta

Figure 4

Arrangement of ionization chamber and scintillation counter for compensation for a single gaseous contaminant



energies of the different detectors, this method is suitable if there is only one contaminant.

With small proportional counters [13], the response to the higher energy gaseous beta-emitters can be further reduced by use of a coincidence arrangement. For example, the relative responses to tritium and ^{41}A may be 10:1 or 5:1 for ^{85}Kr, but the effectiveness of the cancellation is dependent upon the energy of the interfering betas.

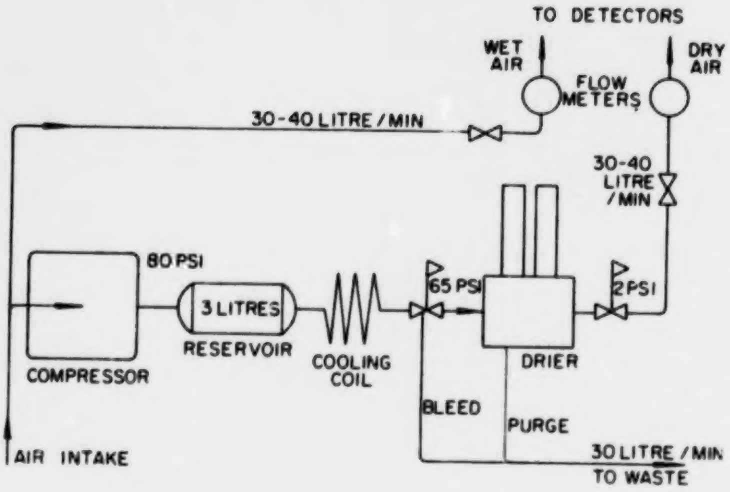
With the plastic scintillation detector, pulse height analysis allows discrimination in favor of tritium but adsorption of water on the scintillator results in the sensitivity to tritiated water vapor being dependent upon the relative humidity of the sampled air.

Method for Continuous Removal of Tritiated Water Vapor from Air

Where high levels of interference are to be expected, a practical solution, in the case of monitoring for tritiated water vapor, has been to measure samples before and after removal of the water vapor, either sequentially with one detector or in parallel with two detectors [11,12, 18]. In the latter case, detectors with identical geometries are used. Various desiccants such as molecular sieves have been used. The problem with this approach is that desiccants have to be renewed, preferably before they are saturated; operating modes in which the desiccant is only switched in when the output signal from the tritium detector increases only reduces the frequency of changes if the "normal" situation is no activity. An alternative to desiccation is to wash the tritiated water from the sampled air. Since this would leave the air saturated with moisture (unless a drying stage were included), which is very undesirable in ionization chambers, desiccation appears preferable for

Figure 5

Continuous drying system for removal of tritiated water vapor from air passing to one of a pair of detectors



use with this type of detector.

To attain continuous tritiated water vapor removal, a two-column cycling sieve drier has been used ("Puregas" Heatless Dryer HF 200-109-1). Figure 7 shows the arrangement of components. The flow of sample to be monitored is divided into two, one part going directly to a detector, the other part being compressed and dried at high pressure in the drier. The pressures at the drier input and output are regulated. Flow rates to both detectors are controlled and metered. The drier operates continuously, the two columns alternating in drying and being dried on a 60-second cycle. An interlock and pressure switch safeguards the compressor against blockage at the input. The dew point at the output from the drier is less than -50°C at the flow rate (500 to 600 cm/s) used, so that the residual tritiated water vapor is less than 0.2% at an input dew point of 10°C. With two 40-liter ionization chambers (gamma-compensated as described in reference 15) connected "back to back," currents from noble gases can be cancelled to less than 1%. The relative responses to tritium and ^{41}A, for example, are more than 20:1. At room temperature, the binding of the noble gases by the molecular sieve is small so that the response time of the system is determined mainly by the total volume and flow rate. The overall response time of the system is 85 s. With our present design of gamma-compensated ionization chamber, in which the compensating chamber is inside the ventilated chamber, the former cannot be used for noble gas compensation. Although the volumes are equal, the geometries are different. However, with the rotating, identical geometry system [16], such dual use (gamma + noble gas) of the compensating chamber is possible though mechanically difficult. With smaller detector volumes or higher flow rates, faster responses obviously could be attained than that noted here, the latter being more difficult from a practical point of view. The advantages of this approach with ionization chambers are that the complete monitor is mechanically and electrically simple and robust. A similar drying system could be used with the more sensitive proportional counter but at the expense of additional complication.

Methods Involving Conversion to the Liquid Phase

An alternative method to eliminate the interference from noble gases is to measure tritiated water vapor in the liquid phase. Provided the volume of water into which the tritium is collected is small compared with the volume of sampled air, the tritium is very well separated from the noble gases and any of the techniques for measuring tritiated water may be used. Some of the possibilities for changing to the liquid phase and the sensitivities for tritium in air that can be attained are noted below.

Although direct condensation of the tritiated water vapor and subsequent detection of tritium in a small scintillation flow cell has been demonstrated [19], this method has the disadvantage that the specific activity of the tritiated water in the air is measured, rather than the concentration per unit volume of air; i.e., the sensitivity is dependent upon the dew point of the sampled air. A way around this is to bring the dew point up to some preset value before condensation, but this leads to a fairly complicated piece of engineering.

A simple but very effective way to change to the liquid phase is to use a water-filled gas washing bottle (or "bubbler"). The activity in the water collected from a known volume of air is measured, usually with liquid scintillator. For a practicable range of flows and

Figure 6

Arrangement of exchanger and detector for the continuous sampling of tritiated water vapor from air and subsequent measurement in the liquid phase

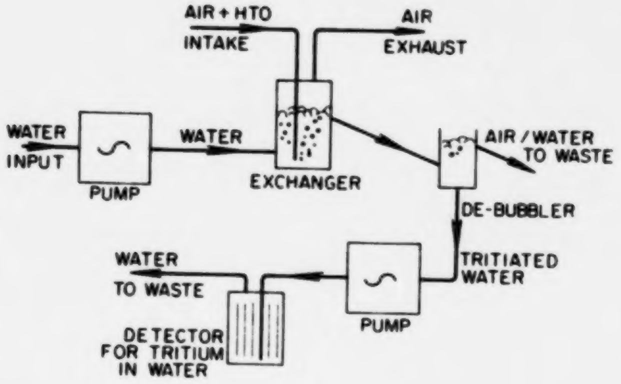


Figure 7

Response of detector, attached to continuous air sampler, to a 45-minute pulse of tritiated water vapor at the sampler intake. Plastic scintillation detector II was used in a Beckman Beta-Mate II scintillation counter. Water flow rate was 0.21 ml s^{-1} through 160 ml exchanger; 0.072 ml s^{-1} through 16 ml detector. Air flow rate was 100 ml s^{-1}.

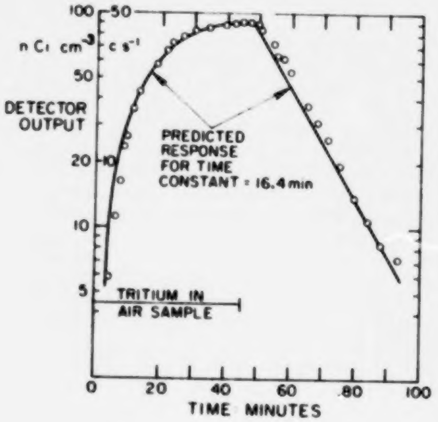
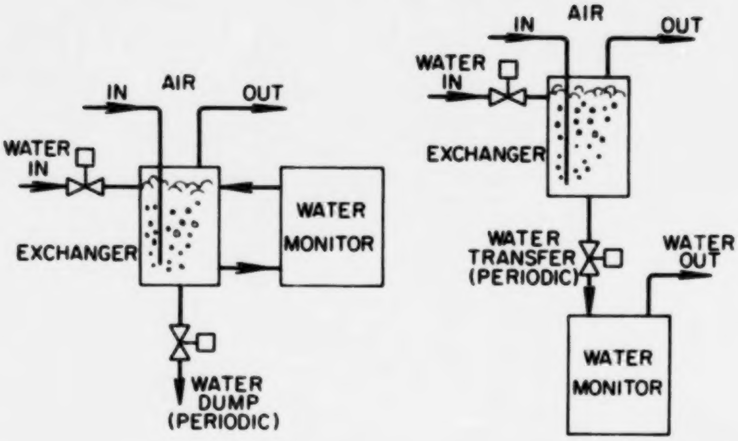


Figure 8
Outline of alternative air sampling systems



flow rates, the collection efficiency of normal laboratory bubblers is calculable provided that the input air, humidity and water temperature are known [20]. However, it is unnecessary to measure these in practice; a simpler approach is either to use a second (or more) bubbler(s) in series with the first, estimating the total activity passed into the bubbler chain from activities collected in the second and subsequent ones, or to sample in such a way that the overall collection efficiency is within a few percent of 100%, irrespective of input or output humidities. For example, sampling at 10 ml/s for 10 hours (or 100 ml/s for 1 hour) through 200 ml of water has an overall collection efficiency of 96 to 97%. The minimum detectable activity in this case would typically be 0.1 pCi ml. In this example, the ratio of noble gas activity to tritium activity in the water would be <0.013% of the average of the ratio in the sampled air. Hence, the exchanger method gives independence from humidity variations and noble gas radionuclides but at the expense of sensitivity.

An attractive possibility is to use this exchange method on a continuous basis in conjunction with one of the tritium-in-water detectors described previously. Many arrangements are possible; a simple one is shown in Figure 6. Water passes from the exchanger to the detector, the flows being arranged so that air is not included in the water flow to the detector. The counting rate in the detector is proportional to the concentration in air. The response of such a sampling system to a step input of tritiated water vapor to the air intake is shown in Figure 7.

Other possibilities noted in Figure 8 include: A circulating exchanger/detector system, the contents of which are periodically dumped (the counting rate in this case is proportional to the product of concentration and time); or a bubbler exchanger which is emptied into a detector periodically (here the counting rate is proportional to the average concentration during the last sampling period). Despite the variety of arrangements, general limitations can be deduced.

If
C = concentration of tritium in air - μCi/ml
S = concentration of tritium in water in the detector - μCi/ml
E = collection efficiency of tritiated water vapor for the exchanger used
V = volume of air sampled - ml
W = volume of water in detector and exchanger containing activity from volume of air V - ml

Then

$$S/C = EV/W \tag{1}$$

If, in a flow exchanger
F = air flow rate - ml/s
U = water flow rate into the exchanger - m/s
H = humidity in the incoming air - ml/ml

Then

$$S/C = E\,F/(U + E'FH) \tag{2}$$

where E' is the collection efficiency for water vapor. For the present purpose the isotope effect can be ignored and E' taken as equal to E.

For high sensitivity S/C should be as high as possible. However, for S/C to be insensitive to variation, δH, in the incoming humidity,

$$EF \ll U/\delta H \tag{3}$$

In addition, the response time (T s) of the sampling system will be not less than W/U, so, for a given maximum response time,

$$U \gg W/T_{max.} \tag{4}$$

Expressions (2), (3) and (4) are general conditions for any exchange system. To illustrate with a particular example, if the plastic scintillator detector (volume 16 ml), described earlier, is used with a 100 ml exchanger for sampling air which may range from very dry to 25°C dew point and for which a time constant no longer than 600 s is required, then, from (4)

$$U \gg 0.2 \text{ ml/s}$$

and from (3)

$$EF \ll 10^4 \text{ ml/s}$$

Allowing up to approximately 5% inaccuracy from humidity changes,

$$\mathrm{EF} \le 500 \text{ ml/s}$$

From (2)

$$(S/C)_{max} = 2500$$

Hence if the minimum detectable activity in the water is 5 nCi/ml the minimum detectable activity in air (C_{min}) is 2 pCi/ml. With a laboratory type of bubbler of this size, the upper limit to EF is about 100 to 200 ml/s so that C_{min} would be 5 to 10 pCi/ml. C_{min} equal to 2 pCi ml would be attained with a response time of 3000 s. However, as Stephenson [21] has pointed out, and as appears from mass transport theory [22], very much higher rates of transfer of tritiated water across a vapor/liquid boundary are possible so that the condition in expression (3) may be the limiting one in a well-designed exchanger. Obviously, by substituting a more sensitive detector for tritium (such as one using liquid scintillator), concentrations of tritium below 1 pCi/ml could be detected but with the concomitant disadvantages of a liquid scintillation system (complication, expense) as outlined earlier.

A sensitivity as high as with direct condensation, but without the humidity dependence, can be attained [20] by bubbling directly through dioxane-based liquid scintillator. This is similar to a method proposed some years ago for chromatographic purposes [23]. Water can be collected in this way up to the miscibility limit of the scintillator. So far, this method has only been tested in the laboratory.

Conclusions

For measuring tritium in water effluents, a plastic scintillator detector offers a fast response (several minutes) with a capability of measuring down to 1 nCi/ml. Radionuclides at levels comparable to those of tritium have to be removed from the samples to avoid serious interference with the measurement of the tritium. Greater sensitivity and discrimination against other nuclides are attainable with liquid scintillator. This monitoring technique has been automated but running costs are high for other than very long response times (hours).

For measuring tritium in effluent air, several kinds of monitors have the inherent sensitivity needed. As with water effluents, one of the main problems is discriminating against other radionuclides. For use with ionization chambers monitoring for tritiated water vapor, a subtraction method using a self-renewing drier has been developed. The system is inexpensive and robust and can have a short response time (minutes). An alternative approach is to use a water exchanger to separate the tritiated water vapor from interfering nuclides, the tritium activity being measured with a scintillation detector. This has a higher discrimination against the noble gases but for measurement at pCi/ml levels (in air) with two phase detectors (solid/liquid), long response times are needed. Liquid scintillation detection appears practicable, allowing much higher sensitivity and/or faster response times, but is expensive.

Table 2

Methods for Effluent Monitoring

Effluent	Requirement	Detection System
Water	high sensitivity (pCi/ml)	liquid scintillator, automatic
	fast response (minutes)	plastic scintillator
Air	high sensitivity (pCi/ml - fCi/ml)	water exchanger with plastic scintillator (or liquid for highest sensitivity)
	fast response (minutes)	ionization chambers (with drier)

Table 2 summarizes the possibilities that appear most appropriate to the types of measurement noted in the opening paragraphs of the paper.

ACKNOWLEDGMENTS

I am grateful for the advice and encouragement of Mr. G. Cowper and the assistance of Messrs. A. S. Coveart and N. W. Tepley, in developing the various ideas, methods and devices described in this paper. I also acknowledge the cooperation and helpful comments of Messrs. P. A. Coyne, A. D. Ironside, and R. K. Mohindra at the various Ontario Hydro Nuclear Power Stations, and of Mr. R. Wilson and members of the Medical Services Division of Ontario Hydro.

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LABORATORY SYSTEM FOR TRITIUM ANALYSIS OF LARGE NUMBERS OF ENVIRONMENTAL SAMPLES

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The National Environmental Research Center, Las Vegas, performs tritium analysis on a variety of environmental samples. The Laboratory surveillance activities result in the collection and analysis of samples of surface water, atmospheric moisture, milk, urine, vegetation, soil, blood, animal tissue, and natural gas.

Techniques were developed for sample preparation and analysis of large numbers of samples. These techniques can be performed routinely by laboratory technicians. Quality control and calibration techniques were incorporated with the counting procedures to assure accuracy and precision of the data. Computer programs were written to perform calculations and generate necessary reports.

Atmospheric Moisture

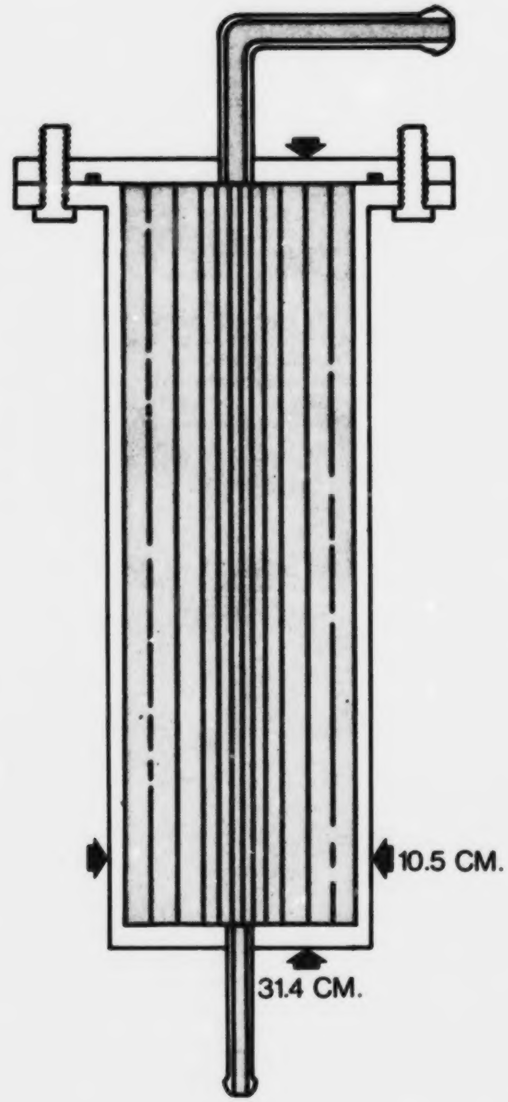
The routine determination of tritium in atmospheric moisture necessitated the development of apparatus and procedures for removal of water from the Linde 13X molecular sieve which was used by surveillance personnel to collect atmospheric moisture. The Linde 13X molecular sieve was selected because it will efficiently remove water and carbon dioxide from the atmosphere [1].

Three types of molecular sieve samplers are used to collect atmospheric moisture. A 300-g sampler is used for portable ground level monitoring, a 600-g sampler for monitoring at fixed ground stations over a 48-hour period, and a 1000-g sampler for collecting atmospheric moisture during aerial monitoring.

One type of degasser is used to remove the adsorbed moisture from the 600-g and 1000-g molecular sieve samples and a second type for the 300-g samples. The 600-g and 1000-g molecular sieve degassers were fabricated from aluminum. The bodies were constructed from 8.9-cm I.D. aluminum tubing, and the bottom plates, top flanges, and cover were constructed from 0.95-cm aluminum plates. The tops were sealed with 0.32-cm by 10.8-cm by 11.4-cm Teflon 'O' rings. Brass 18/9 ball joints were used for the inlets and outlets. The degasser is illustrated in Figure 1. Degassers are not used to collect samples due to the cost of fabricating a large number of degassers and their weight and bulk.

Transferring the molecular sieve from the molecular sieve container to the degasser exposes the molecular sieve to ambient air for only two to five seconds. This exposure does not cause any detectable contamination. The molecular sieve exhibits memory effect when the specific activity of the water is in the microcurie range; however, any memory effect is avoided by discarding it after one usage.

Figure 1



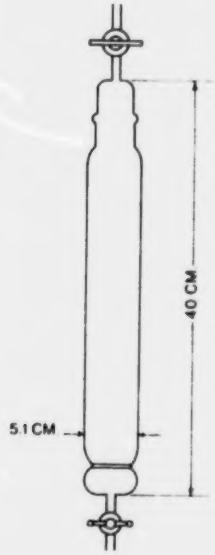
The procedure for removing water from 600-g to 1000-g samples consists of loading the molecular sieve sample into the degasser, connecting the helium line, vacuum trap and vacuum line, and applying heat.. Helium flow is adjusted to 1 liter per minute. The temperature is allowed to rise to 350°C and is held at this temperature for 30 to 45 minutes, which is adequate to remove the water.

The time required to simultaneously remove water from samples of 600-g or 1000-g molecular sieves is approximately 3 hours. Water recovery is 99 percent if the original degassing temperature and the water removal degassing temperature are within ±5°C.

The procedure for removing water from samples of 300 g of molecular sieve is similar to that for 600-g to 1000-g samples except only 10 to 15 ml of water are collected, usually before the temperature reaches 350°C. To remove 10 to 15 ml of water simultaneously from eight 300-g molecular sieves requires 0.5 to 1 hour. Removing only a portion of the water from the 300-g molecular sieves does not cause any detectable isotope effect. The isotope effect is known to decrease at high temperatures.

The absolute humidity of an air sample that passes through a 600-g or 1000-g sampler is calculated from the total quantity of water removed from the molecular sieve and the total air volume. The absolute humidity of air that passes through the 300-g sampler is calculated from psychrometric data collected during sampling. Degassers used to remove water from 300 g of molecular sieve were constructed from Pyrex glass, see Figure 2.

Figure 2



The temperature of the heaters is controlled with variable transformers and monitored with thermocouple gauges.

Water that is removed from molecular sieves is collected in 28- by 200-mm separable vacuum traps. These vacuum traps are immersed in a Dewar flask containing dry ice and acetone.

Procedures used to remove water from 600 g to 1000 g of molecular sieve and those for the 300 g of molecular sieve are different due to the requirement of removing all of the water from the 600-g and 1000-g samples. The water removed from 300-g molecular sieves only needs to be an amount sufficient for tritium analysis. Water samples collected from the molecular sieves are distilled prior to preparation of samples for liquid scintillation counting, to remove any fine particles.

Soil, Vegetation, Animal Tissue, and Blood

Water is removed from soil, vegetation, animal tissue, and blood by vacuum distillation. Vacuum distillation was chosen as the method because it is more rapid and convenient than freeze drying, and isotope effect is less severe at higher temperatures.

The moisture content of soil is determined by oven drying 25 g of a sample at 105°C for 24 hours; for vegetation, 5 g of a sample is dried at 65°C for 24 hours. The moisture content of animal tissue is determined by removal of the water through a 24-hour vacuum distillation of 25 g of the sample. The tissue is cut into small pieces prior to vacuum distillation.

Ten to twenty ml of water are vacuum distilled from soil and vegetation samples for tritium analysis. The time required for vacuum distillation of six vegetation or soil samples is approximately 1 hour.

The water removed from soil, vegetation, and animal tissue is collected in 28 mm or 200 mm separable vacuum traps at liquid nitrogen temperature.

Dow-Corning antifoam emulsion H-10 is added to blood prior to distillation to prevent foaming. To distill water from a blood sample requires approximately 1 hour. Water that is removed from soil, vegetation, animal tissue, and blood is oxidized with potassium permanganate to destroy any volatile organics that distill over with water. The water is then distilled.

Milk, Water, and Urine

Water is removed from milk and urine by distillation, using standard micro glassware. Urine samples are oxidized with potassium permanganate prior to distillation to prevent volatile organic constituents from distilling over with the water. The organic constituent may cause chemiluminescence of the scintillation solution. Dow-Corning antifoam emulsion H-10 is added to milk and urine before distillation to prevent foaming.

Simultaneous distillation of 4 milk, water, or urine samples requires approximately 15 minutes.

Natural Gas

Natural gas samples are combusted by passing the gas over copper oxide at 750°C. The water is collected in a trap immersed in dry ice and acetone. The water is distilled to remove contaminants and prepared for liquid scintillation counting.

The Preparation of Samples for Liquid Scintillation Counting

Five ml of the previously distilled sample and 20 ml of scintillation solution are pipetted into a 25-ml polyethylene vial. The scintillation solution is made from 8 g of 2,5-diphenyloxazole (PPO), 1.5 g of p-bis-(o-methylstyryl)-benzene (bis-MSB), and 120 g of naphthalene made to 1 liter with p-dioxane [2]

Liquid Scintillation Counting

Four Beckman LS-100 ambient temperature counting systems with special low-background photo-multiplier tubes are used for counting. Samples are placed in the counters and allowed to dark adapt for at least 5 hours. A tritium standard and background are counted after every tenth sample. Typical background and counting efficiency are 6 cpm and 23 percent, respectively.

The estimated minimum detectable activity using a 100-minute counting time is approximately 400 pCi/l of water. The minimun detectable activity is defined as the point where the net count rate for the sample and the 2σ counting error are equal. The four-counter system is capable of counting 40 samples per day.

Quality control charts are maintained on each of the counters and are updated daily. If the background and standard count rate exceed $\pm 2\sigma$ of their mean value, the counters are checked for electronic problems and the samples are recounted. Quality control cross-check standard samples and duplicate analysis samples are counted routinely on a monthly basis.

Gross counts per minute and the counting time in minutes for each sample are produced by a printer on each counting system. Counting and sample control data are transferred to a computer coding sheet. The computer produced a preliminary report which is used to check the data and results for errors. Computer result cards are returned to the computer section for production of a final report.

These systems are capable of analyzing tritium in water, atmospheric moisture, milk, urine, soil, blood, animal tissue, and natural gas. Forty samples a day can be analyzed with an estimated minimum detectable activity 400 pCi/l of water. All operations described are routinely run by technicians.

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monitoring stations in the environment of the following reactors (as numbered in Figure 1):

1. Peach Bottom Atomic Power Station
2. Limerick Generating Station
3. Three Mile Island Nuclear Station
4. Calvert Cliffs Nuclear Power Plant

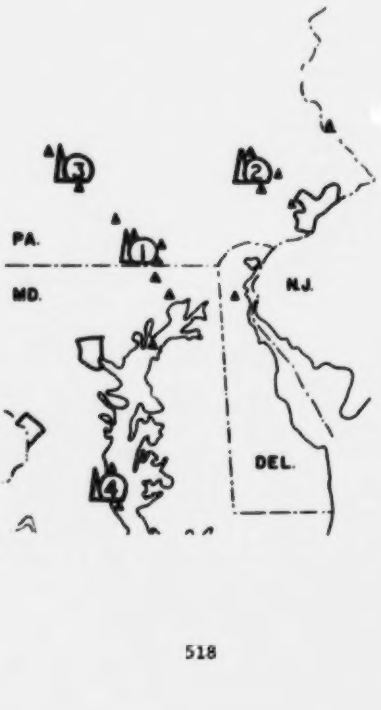
The last two plants have been recently added to the program: thus, no data are presently available for them.

Sample Preparation and Counting

Each sample is first distilled under vacuum to remove dissolved gases and bioluminescent materials. For a minimum detectable activity (MDA) of 400 pCi/l, the sample is dark-adapted and counted in RMC's low-level liquid scintillation system. The discriminator and gain settings of RMC's Packard Tri-Carbs have been carefully modified and manipulated [5]. These liquid scintillation counting systems for a 2-ml sample have a background of 13 cpm and 30 percent efficiency. (A second channel is set wide to detect any other β or γ emitters that would produce Compton scatter in the tritium channel.) The sample is normally counted for 800 minutes, utilizing a cocktail containing 2-phenyl-5-(4-biphenyl) oxadiazole (PBD) in a concentration of 8 g/l of one-third Triton X 100 and two-thirds Toluene. A total sample preparation time and counting time of 830 minutes produces a MDA of 400 pCi/l.

Figure 1

RMC Regional Tritium Monitoring Network



For an MDA of 10 pCi/l, or three tritium units (TU), the sample is processed in the RMC Electrolytic Isotopic Enrichment System built according to Ostlund [6] and Metson [7]. The electrodes are Ni and stainless steel and the glass cells have a 100-ml capacity with an automatic liquid feeding attachment.

Quality Control is an important part of any monitoring program. Each series of samples processed at RMC also contains duplicate spiked samples and blank samples to verify the techniques, in addition to the internal standard in the liquid scintillation counter. Intercomparison samples (split samples) are analyzed in conjunction with the Pennsylvania Department of Environmental Resources, Office of Radiological Health, Environmental Monitoring Branch, the U. S. Environmental Protection Agency, Analytical Quality Control Service, N. E. Labs, and the U. S. Department of Interior, Water Resources Division, Geological Survey, Tritium Lab.

Results

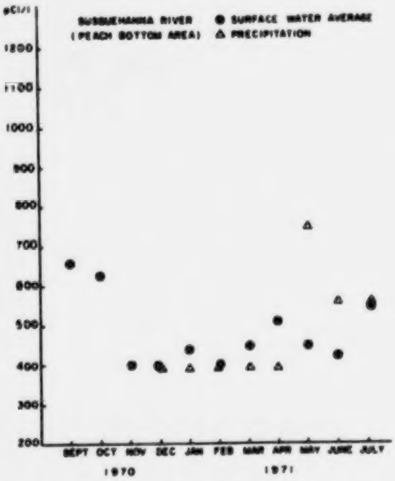
Susquehanna River

From September 1970 through July 1971, the average tritium concentrations in the Peach Bottom Reactor area were as follows:

Surface Water	480 ± 190 pCi/l	(Avg. of nine locations)
Well Water	470 ± 190 pCi/l	(Avg. of three locations)
Precipitation	480 ± 260 pCi/l	(Single location)

Figure 2 indicates the individual variations of surface water and precipitation from month to month.

Figure 2



Schuylkill River

The Limerick Generating Station environmental waters were sampled from February through July, 1971. The results of the five-month average (no April sample) are as follows:

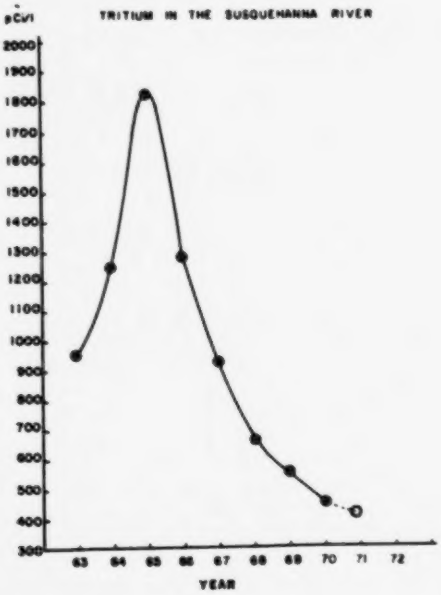
Surface Water	450 ± 210 pCi/l	(Avg. of six locations)
Well Water	< 400 pCi/l	(Avg. of two locations)
Precipitation	710 ± 390 pCi/l	(Composite of five locations)

Long Term Tritium Levels

Data gathered from the U.S. Department of Interior Tritium Lab [8], the Public Health Service, and the more recent RMC data, indicate that the surface waters of the Susquehanna River (from 1961 to present) peaked to about 2000 pCi/l in the mid-1960s (due to weapons testing). The river tritium levels fell rapidly until 1968-1969, then slowly dropped to about 300 to 500 pCi/l at present.

Figure 3

Tritium in the Susquehanna River



Conclusions

Many samples are presently below the level at which they can be directly measured without elaborate sample preparation and/or sample concentration (~400 pCi/l). For nuclear facility environmental monitoring programs, it is recommended that every three months a composite of like samples (such as surface water) be counted to an MDA of 50 pCi/l so that the actual activity will be known. This data (precipitation, surface and well water) should be analyzed to calculate the tritium inventory and its movement through the regional hydrosphere utilizing precipitation rates and river flows.

It is suggested that a comprehensive tritium sampling program be initiated at least two years prior to reactor start-up. This should give the utility an idea of seasonal variations of both surface water as well as the precipitation input. Continuous composite samples should be taken whenever practical.

A coordinated program should include sampling of the local environments of all major tritium producers or users (power reactors, fuel reprocessing plants, tritium dial painting facilities). This program should be coordinated with the local state health departments and the Environmental Protection Agency Monitoring Network.

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ANALYSIS FOR TRITIUM IN WATER: INTERCOMPARISON STUDY OF NOVEMBER 1970

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Tritium has become a radionuclide of concern to the Environmental Protection Agency, U.S. Public Health Service and other government agencies. Due to the advent of nuclear power, tritium is being introduced into the environment in greater quantities. Many Federal, State, and private agencies have begun analysis of this nuclide on a routine basis to measure the buildup in the environment.

A procedure of quality control to assist laboratories in producing accurate and comparable data on tritium concentrations in water is necessary. The Analytical Quality Control Service (AQCS) has attempted to aid this endeavor. In addition to conducting an interlaboratory tritium cross-check program, the AQCS conducted this intercomparison study in November 1970, to assist the laboratories in evaluating their precision and accuracy. Three different levels of tritium were used, including a biological sample, to study the response of tritium radiochemical techniques as used by each laboratory. These levels are above those normally found in the environment so that the laboratories may isolate, identify, and correct any problems associated with their analyses. One problem that arose was unforeseen. The activity of the spiked solution was four percent low from that originally reported. This was first detected by observing that the data from all the laboratories seemed to be biased low.

Results of this study were compared to the established deviation of 10% (for values above 5 nCi/l). This value was determined by examination of data from an AQCS cross-check tritium program which has been in operation for the past three years. The EPA Laboratories and three other government agencies are participating in that program. It was recently confirmed by the previous study performed on tritium [1].

The experiment by the laboratories consisted of triplicate analyses on one biological (urine) and three one-liter distilled water samples (Samples A, B, and C) containing known concentrations (not disclosed to the laboratories) of tritium. The biological sample (Sample D) was prepared by adding a calibrated solution of tritium to a liter aliquot of urine obtained from random laboratory personnel at NERHL. The urine was preserved with thymersol.

The laboratories received 50 ml of each sample, and an unspiked portion of distilled water was supplied for background determination. Each laboratory is identified in this report by a letter to maintain the confidentiality of the results.

Figure 1

Examples of Mean and Range Control Charts
Mean Chart - Sample A. ^3H = 28.1 nCi/l N = 3

$\overline{X} + \frac{3\sigma}{N}$	33.0	Upper Control Limit (UCL)
$\overline{X} + \frac{2\sigma}{N}$	31.3	Upper Warning Level (UWL)
$\overline{X}$	28.1	Standard Mean (Expected Value)
$\overline{X} - \frac{2\sigma}{N}$	24.9	Lower Warning Level (LWL)
$\overline{X} - \frac{3\sigma}{N}$	23.2	Lower Control Limit (LCL)

Range Chart - Sample A. ^3H = 28.1 nCi/l N = 3

$D_4\overline{R}$	12.1	Upper Control Limit (UCL)
$\overline{R} + 2/3\ (D_4\ \overline{R}-\overline{R})$	9.6	Upper Warning Level (UWL)
$\overline{R} = d_2\sigma$	4.7	Mean Range (Expected Range)
$D_3\ \overline{R}$	0	Lower Limit (LL)

Figure 2

Mean Chart Sample A 26.8 nCi/l N = 3

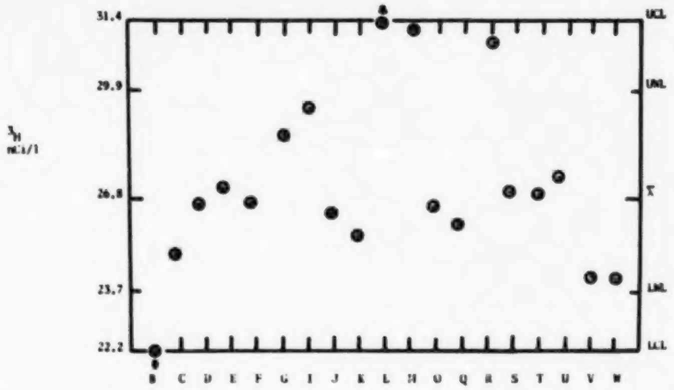


Figure 3

Range Chart Sample A 28.1 nCi/l N = 3

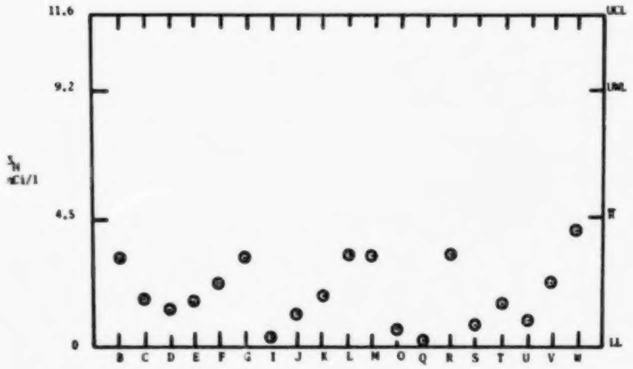


Figure 4

Mean Chart Sample B 50.5 nCi/1 N = 3

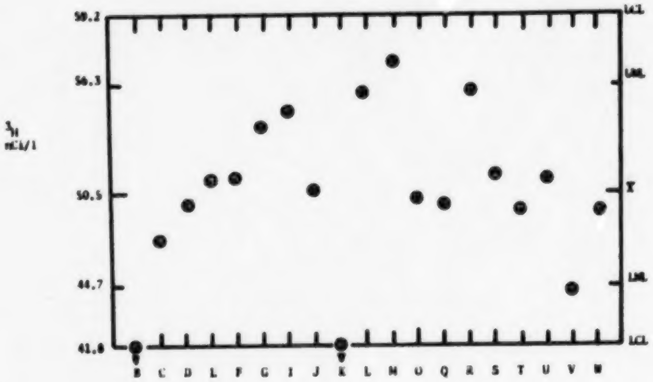


Figure 5

Range Chart Sample B 52.9 nCi/1 N = 3

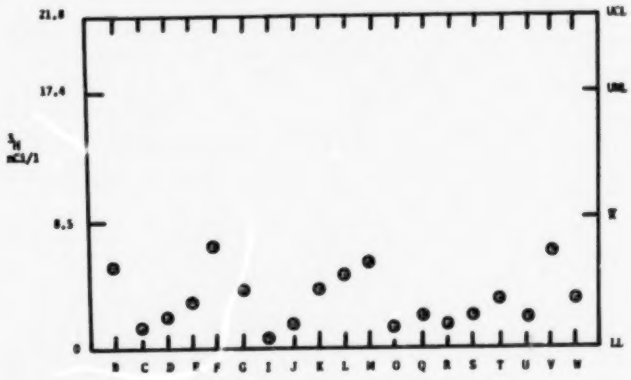


Figure 6

Mean Chart Sample C 133.8 nCi/l N = 3

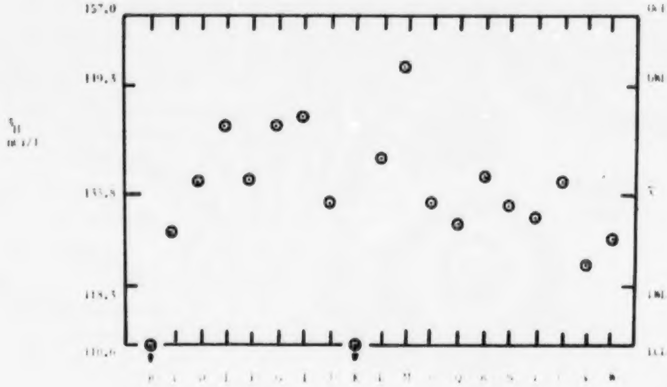


Figure 7

Range Chart Sample C 140.3 nCi/l N = 3

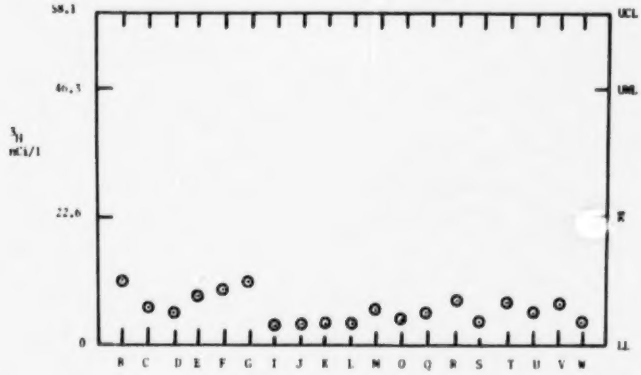


Figure 8

Mean Chart Sample D 55.1 nCi/1 N = 3

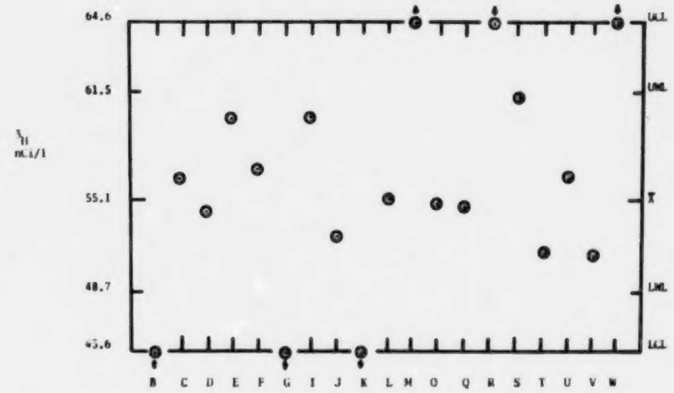
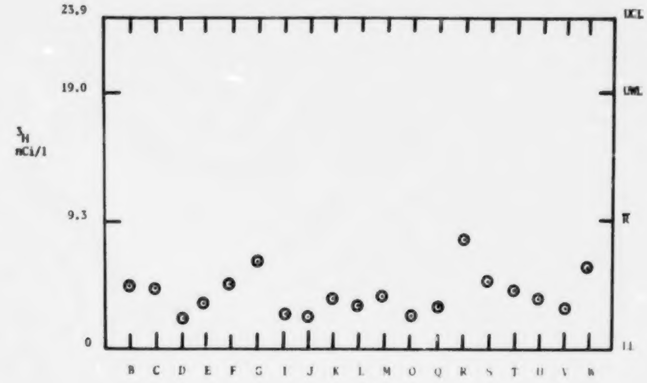


Figure 9

Range Chart Sample D 57.8 nCi/1 N = 3



Data Analysis

The data were statistically analyzed with the aid of control charts (Figures 2-9) to determine if each of the individual laboratories met the proposed standards of internal precision and analytical agreement. This method of analysis has been described elsewhere [2]. Tables 1 and 2 present the distribution of laboratory mean and range values at the various control levels.

Table 1

Distribution of Mean Results
Number of Laboratories in Each Category

^{3}H, nCi/l	$\leq\overline{X}$ ± WL	$\leq\overline{X}$ ± CL	$>\overline{X}$ ± CL	Total
26.8	15	2	2	19
50.5	15	2	2	19
133.8	16	1	2	19
55.1	13	0	6	19

Table 2

Distribution of Mean Results [1]

^{3}H, nCi/l	$\leq\overline{X}$ ± WL	$\leq\overline{X}$ ± CL	$>\overline{X}$ ± CL	Total
51.8	9	3	4	16
146.4	9	1	6	16
331.4	9	3	4	16

Two calculations are made on each set of results. The mean and the range of the three determinations are calculated. When the number of determinations is small (as in this case), range is almost as useful as standard deviation as a measurement of precision.

The mean and range values obtained are plotted on two types of control charts as illustrated in Figure 1. The mean control charts determine whether the degree of bias in the data is excessive, and the range charts determine whether the precision within a single laboratory is acceptable.

The following nomenclature is used in Figure 1:

$\overline{X}$ = the known activity level being studied.

$\overline{R}$ = the expected value of range, i.e., the average value that would be observed if a very large number of range determinations were made under acceptable conditions.

σ = the standard deviation for single determinations.

N = number of replicate determinations (in this case: three).

d_2, D_4, D_3 = values taken from standard statistical tables [2] prepared for the construction of range control charts and based on the value of N.

When N = 3

$$d_2 = 1.69$$
$$D_4 = 2.57$$
$$D_3 = 0$$

Results and Conclusions

The results of this experiment have been analyzed statistically by means of control charts. The control chart for means enables a laboratory to measure the amount by which its mean value differs from an expected value. It is assumed that under satisfactory conditions data would fall within the limits of the control chart, and if a particular mean fails to meet the limits, it would be considered unacceptable. The control chart for ranges enables a laboratory to measure its own precision. If a particular range value does not meet the limits of the control charts, there is an indication that the laboratory's precision is unacceptable.

For a laboratory to evaluate its performance in this experiment, it should locate the position of its mean and range values for a particular determination on the appropriate control chart. Results are completely acceptable if the mean and range values fall within the warning levels for each concentration of the particular radionuclide which was measured. Results are conditionally acceptable if the mean and range values fall within the control limits, but the laboratory should consider ways to improve the determination. Values which fall outside the control limits indicate that action should be taken to investigate the reason for failure to meet the control limit. The various levels of radioactivity which were studied help each laboratory to uncover any problems which exist. High levels detect major calibration and procedural errors, and intermediate and low levels detect errors due to instrument response, electronic stability, reagent blank and counter background.

In general, the mean results show considerable improvement over previous experiments. When compared with the results from the previous experiment [1] given in Table 2, it can be seen that the number of laboratories outside the control limits (19%) are less even though three more laboratories took part in this experiment. The values for the urine samples were especially encouraging since some of the laboratories do not routinely analyze this type of media.

Precision remains good as can be seen on Table 3. All analyses fell well within the Upper Warning Level for all samples including the biological sample (55.1 nCi/l). This shows that the laboratories' reproducibility is good. A first check on the data showed a low bias. Many of the laboratories participating in this experiment had obtained standards from the NBS on an independent basis. Therefore, this low bias could not be due solely to the methodology, since each laboratory uses its own procedure in these experiments.

However, a least squares determination indicated that either the laboratories were biased low or that the standard used was higher than its true value. A thorough investigation confirmed the new value.

Table 3

Distribution of Range Results
Number of Laboratories in Each Category

^{3}H, nCi/l	$\leq\overline{X}$ + UWL	$\leq\overline{X}$ + UCL	$\overline{X}$ + UCL	Total
26.8	19	0	0	19
50.5	19	0	0	19
133.8	19	0	0	19
55.1	19	0	0	19

The original values for the known samples are listed and are compared with the later confirmed values (Table 4).

Table 4

Tritium Concentration of Water Samples Used in this Study

Sample	^{3}H, nCi/l (original)	^{3}H, nCi/l (verified)
A	28.1	26.8
B	52.9	50.5
C	140.3	133.8
D	57.8	55.1

The charts were replotted, and the values check. The values for the laboratories were changed if necessary. Those previously calibrated by this specific standard would have to be recalculated. Results were good, and those laboratories having results within the Warning Level now were normally distributed around the mean. The original values as distributed around the mean (Table 1) did not change significantly. It was interesting to note that this type of experiment could detect a slight error (4%) due to the standard used.

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3. E. L. Grant, Statistical Quality Control, McGraw-Hill, NY, 1964, pp. 561-562.
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A TRITIUM AIR SAMPLING METHOD FOR ENVIRONMENTAL AND NUCLEAR PLANT MONITORING

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The need for detecting tritium air concentrations below prevailing radiation standards was recognized prior to the emphasis on environmental impact. However, tritium sampling methods now in use either require time-consuming effort or do not measure low levels of radioactivity. Desiccants, such as silica gel, provide a reasonable method for collecting atmospheric moisture, but recovery requires a considerable investment of time. Silica gel is being used by other groups to monitor for tritium, but the direct analysis of absorbed moisture on the gel is a new technique. The objectives of our research were to establish moisture collection efficiencies for silica gel, to develop a reliable procedure for directly counting tritiated moisture adsorbed on silica gel, and to relate the tritium air sampling concentration measurements to standardized tritium concentrations under the controlled conditions of an environmental chamber. Even though elemental tritium exists at low altitudes, tritiated water is the most hazardous form of contamination. Therefore, the measurement of tritium oxide air concentrations provides a realistic evaluation of nuclear plant or environmental conditions.

Method

The air flow from most sampling systems can be directed through a column of silica gel to collect atmospheric moisture, or a complete sampler may be assembled. The sampled air is filtered to remove airborne particulates and diverted through a plastic column or tube of silica gel at a regulated flow rate. The column is made of lucite plastic, with a length of 32.4 cm (12.75 in), an outside diameter of 5.1 cm (2.0 in), and an inside diameter of 3.8 cm (1.5 in). A plastic screw cap with an O-ring seal is used on one end of the column and the other end is sealed with a plastic disc. Flanged tube fittings are used for connecting hoses to each end of the column, which will hold 250 g of silica gel. Air flow through the silica gel is regulated by a needle-valved flow rate meter. The calibrated flowmeter controls air flow from 2.8 to 28 l/h.

The sample column used to collect tritium oxide contains a 10 percent blue moisture-indicating silica gel uniformly mixed with 90 percent clear gel. Particle size of the color indicating and clear gel is 6 to 16 Tyler Sieve Standard. The volume of air sampled is one cubic meter. After sampling, the section of gel containing adsorbed moisture (pink to clear) is removed from the column, plus 1 cm of the adjacent blue, and mixed. A 5-g aliquot is taken from the mixed moisturized gel and counted for tritium.

The usual liquid scintillation method for analysis of tritium in water consists of counting 5 ml of water with 18 ml of scintillation solution (PPO, POPOP, napthalene, and p-diozane) for 20 minutes. When moisture adsorbed on silica gel was analyzed for tritium, the method consisted of counting 5 g of gel, 2 ml of water and 18 ml of scintillation solution for 20 minutes.

Results

Table 1 lists the analysis results of several mixtures of a tritiated water standard, distilled water and silica gel, and of the same tritiated water standard without gel. The results show that the presence of silica gel reduced the counting yield by no more than three percent of the normal scintillation counting method. Aliquots of the silica gel with 10 percent moisture-indicating gel can be analyzed for tritium with the same counting yield as for clear gel.

Moisture collection efficiency of the silica gel columns was determined by analyzing gel from different sections of the column for tritiated moisture. Aliquots of moisturized (pink) gel and adjacent dry (blue) gel showed 99 percent of the collected tritium activity was contained in the moisturized gel. The efficiency for collecting atmospheric moisture with silica gel was approximately the same for conditions from 0° to 27°C and relative humidity from 10 to 100 percent. Moisture adsorbed on silica gel in a sealed column did not move into the dry gel when stored for two weeks. The silica gel samples did not indicate gaseous fission product or natural radioactivity being collected by test sampling, but a 24-hour decay period is used to eliminate short half-life naturally occurring isotopes. The analysis procedure also includes counting the silica gel sample for ^{222}Rn, so that activity from longer half-life isotopes is not included in tritium radioactivity results.

Table 1

Results of Analyzing Tritiated Water on Silica Gel and Without Gel

Analyzed Silica Gel Grams (dry)	Standard Tritiated Water ml	Water Added ml	Analyzed Tritium Standard Concentration nCi/ml
5.0	0.10	3.0	0.43
5.0	0.25	2.8	0.46
5.0	0.50	2.5	0.50
5.0	1.0	2.0	0.45
5.0	2.0	1.0	0.45
None	5.0	None	0.47

The analytical limit for measuring tritiated water on 5 g of silica gel is 7 ± 5 pCi, assuming 100 percent error at 2 sigma. Moisture collection efficiency for silica gel is 99 percent. The volume of air sampled is 1.0 cubic meter. The ratio of moisturized silica gel analyzed to the total moisturized gel (in grams) would be 3 for low humidity and 50 for high humidity conditions. Therefore, the detection limit becomes

$$\text{D.L.} = \frac{1.2 \times 10^{-5}\ \text{nCi/5 g (15 g moisturized gel)}}{(0.99)\ (1.0 \times 10\ \text{ml})} = 4 \times 10^{-11}\ \mu\text{Ci/ml}$$

for low humidity air and 6×10^{-10} μCi/ml for high humidity and temperature air.

Discussion

The most efficient tritium air sampling was accomplished with the following procedure: prefiltering the sampled air to remove particulates above 0.8 μm in size, sampling the air through a column of 250 g of silica gel (particle size 6 to 16 Tyler Sieve Standard) with a 10 percent mixture of indicating gel, sampling air at a rate of 6 l/h, weighing the amount of silica gel that was in the region which has turned pink, plus approximately 1 cm, into the blue colored region, and scintillation counting a representative 5-g aliquot of this weighed silica gel. Atmospheric moisture adsorbed on the gel can be counted directly, or the moisture may be recovered for analysis. Directly counting silica gel with absorbed moisture reduces analysis time and effort without sacrificing accuracy. A separate standard of 2 ml tritiated water on 5 g of silica gel, plus 18 ml of scintillation solution, can be used to determine the counting yield for the method and the microcuries of tritium activity per unit volume of air can then be calculated.

* * *

A SAMPLER FOR NONAQUEOUS TRITIUM GASES

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A sampler for combustible forms of nonaqueous tritium gases has been developed as part of a long-term study of the radioactive waste discharges from a nuclear fuel reprocessing plant by the Division of Surveillance and Inspection, Office of Radiation Programs, Environmental Protection Agency (EPA). The sampler has been successfully operated in the laboratory and in field tests at a nuclear fuel reprocessing plant. This sampler was conceived from a similar design developed in 1969 [1] and shares certain design concepts with the sampler used by Ostlund [2].

The nonaqueous tritium sampler employs a flow-through system. Air containing potentially detectable levels of combustible tritiated substances is pumped through a catalytic burner. Employing a platinum on aluminum oxide catalyst, the burner oxidizes these combustible compounds, producing tritiated water. At least one catalyst is commercially available that can bring the hydrogen content of a gas down to about 1 ppmv [2], but the natural hydrogen content of the atmosphere is only 0.5 ppmv. However, by using a carrier hydrogen gas technique virtually all the tritium may be collected, and although the actual H_2 gas concentration is lost, units of tritium activity may be calculated per unit volume of air. The most sensitive commercially available tritium monitors can measure only about 0.5 pCi/ml or 1/2 MPC. Our sampling system can easily be operated to achieve a sensitivity four orders of magnitude lower than MPC.

The burner has been successfully tested in the laboratory with the following hydrogen-containing substances: acetone vapor, denatured ethyl alcohol and hydrogen gas. Sampler efficiency tests based on mass have been performed using hydrogen gas.

Initial field testing was conducted during the summer of 1971 at the Nuclear Fuel Services (NFS) fuel reprocessing plant at West Valley, NY. This plant and its surrounding area have been the site of several radionuclide studies by our laboratory in recent years [4-7]. The fuel reprocessing plant was an ideal location for field application of this sampler because reprocessing plants are an important source of tritium yet the exact character and quantity of tritium effluent has been poorly understood. Released levels of tritiated water vapor as measured are far below levels projected by burnup equations [3]. Prior to first operation of this sampler at NFS, the quantity of elemental tritium gas in the stack was unknown.

A second important reason for application of the sampler to the reprocessing plant is the inherent discrimination property of the sampler. Krypton-85, an inert gas which like tritium is a beta emitter, is many times more plentiful than tritium in the stack at NFS. The use of an ionization chamber, for example, to measure tritium under these circumstances would be impossible. This sampling technique, however, eliminates detection problems caused by the presence of other radionuclides in the NFS stack effluent.

The Sampler and Its Operation

The sampler performs four operations upon the air flowing through it, as shown in Figures 1-3. The first stage consists of mixing the air being sampled with a dilute-prepared mixture of hydrogen in air. This mixture moves on to the second stage, air dryer #1, which removes all water from the stream and acts as a sampler for tritiated water vapor. The third stage, the catalytic burner, converts carrier hydrogen and any combustible tritiated substances to water. The air sample then enters the fourth stage, air dryer #2, which collects all the water vapor produced in the burner. Later, the water is removed from air dryer #2. The water sample is filtered and distilled, then counted using liquid scintillation.

Air dryer #1 is placed in the line primarily for the purpose

Figure 1

Sampling Train for Total Airborne Tritium

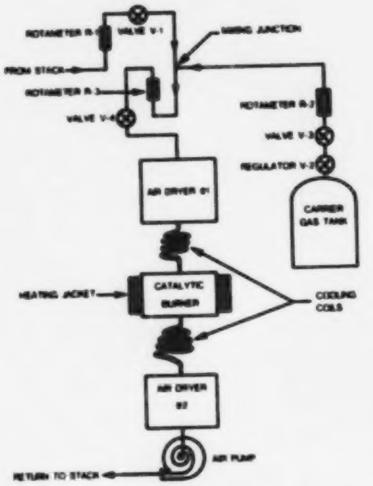


Figure 2

Catalytic Burner

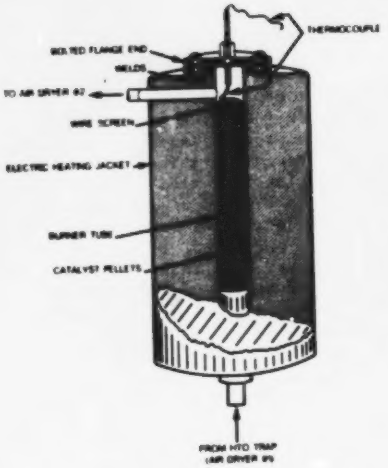
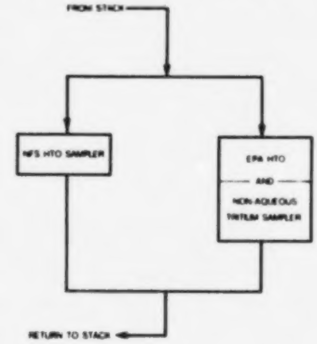


Figure 3

Stack Tritium Samplers at NFS



of removing all tritiated water vapor from the sample air. For this study, Drierite brand anhydrous calcium sulphate obtained from Hammond Drierite Co., Xenia, OH, contained in plexiglas cylinders was used as the air drying agent. A flowing air stream having a temperature of 100°F and 100 percent relative humidity will be dehydrated by Drierite to 5 to 10 ppm by volume. (Moisture content of air is reduced from 45.6 mg/l to 5 μg/l which corresponds to a dew point of -110°F.) When both aqueous and nonaqueous tritium are critical to sampling, air dryer #1 can act as a water vapor sampler. A cooling coil between air dryer #1 and the burner protects rubber and plastic parts from the heat of the burner.

The catalytic burner is probably the most important feature of the sampler. Combustible compounds are oxidized with the help of 0.5 percent platinum on 1/8" diameter aluminum oxide pellets obtained from Engelhard Industries, Newark, NJ. The catalyst is contained in a stainless steel tube, which for our purposes is 12" long X 1" O.D. and fitted with connectors at each end appropriate for connection with the copper tubing coils. The burner tube is surrounded by an electrical heating jacket which maintains a temperature of 400°C in the pipe, catalyst, and air stream.

The temperature at various critical points is indicated by a pyrometer with thermocouples installed in and around the burner. At least one thermocouple is located at the interface between the pipe heating jacket and the stainless steel tube. The temperature at this point should not exceed 500°C, to protect the catalyst [9]. Another thermocouple is installed within the tube near the exit end, and this one measures the temperatures of gases leaving the burner. A temperature of 400°C measured at this point implies that complete combustion has occurred in the burner.

This sampler requires the collection of water vapor at two points in the line. Drierite was used as the desiccating agent for this study; silica gel and molecular sieve have been suggested as methods equally suitable. Cold traps are probably not suitable for the range of flow rates we used, but might be well suited for work at much lower flow rates. Any desiccating method is adequate if properly calibrated for both mass balance efficiency and activity balance efficiency.

$$\text{mass balance efficiency} = \frac{\text{mass of water removed upon dehydration}}{\text{mass of water originally added to desiccant}}$$

$$\text{activity balance efficiency} = \frac{\text{total tritium activity removed in one dehydration}}{\text{total tritium activity originally added}}$$

Drierite, or anhydrous calcium sulphate will absorb 6 to 7 percent of its own weight in water by a hydration process. When heated to near 200°C, the water is released.

Drierite was first calibrated for mass balance efficiency. Using several methods, including liquid phase injection, steam injection, and flow-through of typical room air, masses of water ranging from 2 to 60 grams were used to saturate various quantities. A closed, heated pipe, filled with the Drierite, was heated under vacuum for two hours at 200°C by an electrical heating jacket. Water was collected in a glass trap immersed in liquid nitrogen. Average mass balance efficiency of 1.03 ± 0.05 (1σ) was observed during six runs using 30 to 40 gram samples of water vapor injected into Drierite traps having a capacity of 50 grams water.

Using tritiated water, tests were conducted to determine mass balance and activity balance efficiencies. Even with a mass balance of 1.03, we found an activity balance efficiency of 0.97 ± 0.04 during five runs using the 30 to 40 gram samples of water vapor injected into traps of 50 gram capacity.

The early trials of the catalytic burner duplicated those carried out previously [1]. Acetone vapor was carried in a stream of air through the burner, and the accompanying color change on Indicating Drierite showed that water was produced. The same result was obtained with alcohol.

Two factors were considered when testing the burner. First, the temperature could not exceed 500°C or the catalyst might be adversely affected. Secondly, mixtures of combustible substances entering the burner had to be sufficiently dilute to avoid an explosion. Both of these problems were overcome by using a dilute mixture of combustible gas in air to test the burner.

Three sets of combustion tests were carried out in order to calibrate the burner efficiency for oxidation of hydrogen gas. In each set of tests, the amount of hydrogen injected into the burner was determined by changes in the hydrogen tank pressure. The amount of hydrogen oxidized was determined by measuring the weight increase of the downstream air dryer due to collected water vapor.

$$\text{burner efficiency} = \frac{\text{mass of hydrogen gas oxidized}}{\text{mass of hydrogen injected into burner}}$$

The first set of tests used a commercially prepared and analyzed mixture of 1% hydrogen in air. (This same mixture was also used as the carrier gas for field testing.) During four tests, a steadily flowing stream of 2 l/min of the gas mixture was released into the burner for several hours. The combustion efficiency for this series was 0.98 ± 0.05.

The second set of tests used a pure hydrogen gas source which was diluted to a concentration of 1.5 to 2% hydrogen in air. These mixtures were prepared in our laboratory using a specially constructed apparatus. During ten tests this gas mixture was injected into the burner at a flow rate of 4 l/min. The combustion efficiency for this series of tests was 1.00 ± 0.03.

A third set of nine tests was carried out to investigate whether various concentrations and flow rates have any effect on efficiency. Mixtures ranging from 4 to 7% hydrogen in air were released from the tank into the burner at 2, 3, and 4 l/min. The combustion efficiency resulting from this series was 1.00 ± 0.04. Combustion efficiency was not affected by the stated variations of flow rate and percentage hydrogen.

Field Operation and Testing

Nuclear Fuel Services (NFS) of West Valley, NY, was the site of initial field testing of the sampler from June 2 through June 8, 1971. Combustible tritium gases were found during these short-term tests, so the sampler was operated continuously from August 27, 1971, to November 9, 1971. The sampler for nonaqueous tritium was operated in conjunction with two Drierite traps for water. The source of samples was an existing isokinetic sampling port located at the 80 foot elevation of the 200 foot stack. Connected to the stack port by 210 feet of pipe, the sampling equipment was located in the upper extraction aisle which adjoins the NFS control room. The constantly flowing line has been in use for continuous sampling and monitoring of various radionuclides by NFS for several years. The entire tritium sampling unit in operation at NFS is illustrated in Figure 3.

The NFS water sampler consists of three steel cylinders filled with Drierite and exposed to a flow from the stack of 0.1 l/min. The NFS water sampler operates continuously and is recharged with new Drierite once each week. It is used to find the average weekly stack output of water.

Table 1

Results of Stack Sampling During June 1971 Field Trip

Sampling Description		Sampling Time (min)	Water Recovered (g)	Tritium Activity of Water pCi/ml*	Tritium Activity in Stack Air pCi/ml**
1A	7 hour Pre-dissolution Gas	420	12.3	1.51×10^4	0.154
1B	7 hour Pre-dissolution Water Vapor	420	8.6	7.27×10^3	0.106
2A	Dissolution #1 Gas	496	15.5	2.03×10^5	1.990
2B	Dissolution #1 Water Vapor	496	9.6	9.78×10^3	0.153
3A	Dissolution #2 Gas	482	18.7	2.01×10^5	1.810
3B	Dissolution #2 Water Vapor	482	8.7	1.10×10^4	0.214
4A	Dissolution #3 Gas	1165	48.6	2.56×10^5	0.878
4B	Dissolution #3 Water Vapor	1165	16.0	1.99×10^4	0.208

*1σ due to counting and sample processing ±5%.
**1σ due to counting, processing, and air volume measurement ±8.5%.

Our sampler for water and nonaqueous tritium was operated in parallel with the NFS sampler just described. Upstream of the burner, air dryer #1 removed all water vapors from the stream. The gas sample collected downstream of the burner was later compared with the water vapor sample collected upstream in the water trap, see Table 1. This information was used to characterize total airborne tritium stack effluent at NFS during dissolutions. During the June tests there were four sampling periods. During each period, a water and a nonaqueous sample were collected, making a total of four pairs of samples.

The concept of a dissolution is important in understanding fuel reprocessing and sampling. Sheared fuel rods contained in metal baskets are loaded into large vats called dissolvers. Dissolution begins when nitric acid is injected into the loaded dissolvers. The fission product gases, tritium and krypton-85, which have been trapped within the solid of the fuel elements, are released by the action of the acid. For this reason, stack concentrations of krypton-85 and tritium reach peak levels during a dissolution. Fuel from a given source is normally processed in batches by a series of dissolutions. After the last dissolution is completed, a cleanout operation takes place before work is begun on the next group of fuel. This series of dissolutions on fuel from a single source and the subsequent plant cleanout is referred to as a campaign. Each campaign is named after the source of the spent fuel being reprocessed.

At the time of the June sampling, the plant had been out of operation approximately 30 days, resulting in much lower levels of activity than exists during a routine dissolution campaign. Collection of the first sample pair, defined in Table 1 as "pre-dissolution," came at the end of the 30-day down period and covered the last seven hours prior to the first dissolution. It should be mentioned that two factors may have elevated stack activity levels above the very low levels normally expected after an extended down period. First, the shearing of fuel elements in preparation for forthcoming dissolutions had been in progress for several days. Secondly, the dissolver was completely loaded during the collection of the seven-hour pre-dissolution sample. For these two reasons, this sample may not represent the lowest practical level of activity at the plant.

The air activity shown in Table 1 was obtained by dividing the total activity sampled by the total volume of air passing through the sampler. The air concentrations indicated that most of the tritium gas was evolved during the first eight hours of dissolution, whereas the release of tritiated water vapor was continuous throughout the complete dissolusion cycle.

The June tests proved that detectable levels of tritium are in the NFS airborne effluent and that the sampler described here can be used to detect and measure the non-aqueous tritium, HTO, and total tritium releases. Two more questions merited further investigation: 1) could the sampler be operated continuously over a long period, and 2) how would long-term average releases compare with the short-term data from the June tests? The final period of sampling was 73 days and included two campaigns. One (CVNPA) was 39 days long, lasting from August 27, 1971, to October 5, 1971, and the other (Consumers Power Fuel) was processed from October 5, 1971, to November 9, 1971. Flow rate from the stack sampling line was 0.2 l/m. The sampler operated continuously for 73 days except for the quick changeover from an empty to a full carrier gas tank.

Table 2

Results of Long-Term Stack Sampling

Sampling Description	Sampling Time	Water Recovered (g)	Tritium Activity of Water pCi/ml	Tritium Activity in Stack Air pCi/ml Air
5A Campaign of CVNPA Fuel Gas Sample	8/27 - 1100 to 10/5 - 0930 39 days	149.4	$1.28 \times 10^{4} \pm 5\%$	$1.62 \times 10^{-1} \pm 12\%$
5B Campaign of CVNPA Fuel Water Vapor		210.3	$1.26 \times 10^{4} \pm 5\%$	$2.16 \times 10^{-1} \pm 8\%$
6A Campaign of Consumers Power - Fuel Gas Sample	10/5 - 0930 to 11/9 - 1300 34 days	84.3	$1.18 \times 10^{4} \pm 5\%$	$9.74 \times 10^{-2} \pm 12\%$
6B Campaign of Consumers Power - Fuel Water Vapor		205.3	$5.10 \times 10^{4} \pm 5\%$	$9.97 \times 10^{-1} \pm 8\%$

Campaign Data	Batches	MTU	Burnup MWD	Fuel Cladding Type
CVNPA	10	3.41	34,406	Zircaloy -4
Consumers Power	7	5.93	80,452	Zircaloy -2 and Inconnel

Weekly checks were made on the flow rates and tank pressures, and Drierite traps were removed when full. The data from the two campaigns are in Table 2. For both campaigns the release of HTO vapor was greater than release of tritium gas. This is a contrast to the June tests where, in all four cases, the levels of tritium gas were higher than HTO. Because different fuels were involved, a direct comparison cannot be made between the June results and long-term results. However, it seems safe to conclude that nonaqueous tritium gas output is higher during dissolutions, but that average output of HTO vapor is higher.

The most striking anomaly in the long-term data is the HTO level for the Consumers Power campaign. Note that it is by far the highest HTO level observed of the six HTO vapor samples, despite the fact that it is a long-term rather than short-term sample. We have no explanation for this, but future investigations in the field of fuel reprocessing should clarify the point.

Conclusions

Laboratory and field testing has proven the nonaqueous tritium sampler to be an efficient, simple to operate, durable and reliable device. All substances tested to date have been successfully oxidized in the burner. Its stability and simplicity of operation allow it to be run for long periods of time with little or no supervision; 73 days of continuous operation required only weekly spot checks. It has good reliability because it uses simple mechanical and chemical action instead of complicated electronics which often are temperamental when used in the field. Finally, this nonaqueous tritium sampling capability can be added at small original cost to laboratories already having the capability to measure HTO. The parts of the sampler can be purchased and/or fabricated for less than $300. The costs of operation, for carrier gas and desiccant, are variable and dependent on frequency of operation.

Discussion and Recommendations

The prepared mixture of hydrogen in air need not be used if sufficient combustible gases are available in the sample air. The percentage of hydrogen in air should not exceed the lower limit for an explosive mixture (5 to 10 percent hydrogen in air). The carrier fuel must contain no more than ambient levels of tritium. Use of the carrier gas will dilute the tritium gas, but some carrier is necessary to yield a meaningful sample.

We suggest that rubber vacuum hose not be used for vacuum dehydration of Drierite or distillation because volatile materials released into the sample may cause quenching of liquid scintillation counting. Tygon vacuum tubing is an alternative, because it has better physical and chemical integrity.

Ostlund [2] used a palladium catalyst to oxidize tritium in his sampler. Platinum and palladium have similar catalytic properties. The main difference seems to be that palladium can be operated at lower temperatures.

Note

The mention of commercial products, their source, or their use in connection with materials reported herein is not to be construed as either an actual or implied endorsement of such products by the Environmental Protection Agency.

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A TRITIUM STACK MONITOR WITH A WIDE DIGITAL RANGE

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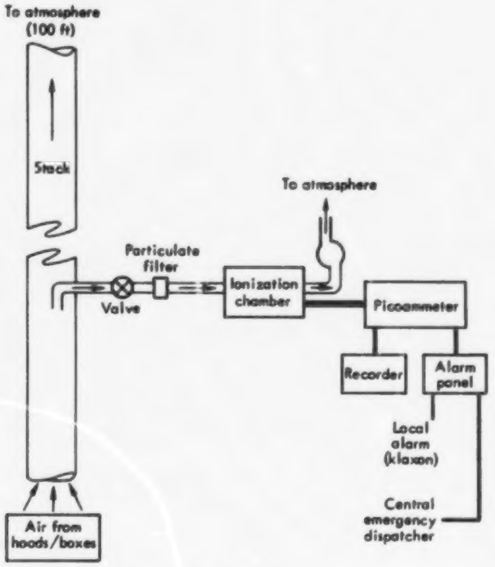
Stack effluent from LLL Livermore's main tritium-handling facility, Building 331, has been continuously monitored for possible tritium discharge since facility construction 12 years ago [1]. The monitoring system (Figure 1) consists of an air mover, a 1-liter ionization chamber, a logarithmic picoammeter, a continuous dual-pen recorder, and an alarm set to trigger at a preset current or release rate. With this system it is possible to measure and record the stack release concentration over a six-decade range from 10^{-4} μCi/ml to 10^{3} μCi/ml. The system has reliably indicated the total tritium effluent. There were, however, two major inherent limitations.

Figure 1



First, in order to determine the total tritium released during a given period, it was necessary to hand-integrate the logarithmic curves and convert ampere-seconds to curies. Although this could be done fairly accurately, there were occasional problems when one quickly tried to estimate the total activity released following an accidental discharge.

The second limitation was related to the problem of alarm setting. The alarm point could be chosen on the basis of concentration only, because the system had no accumulating capability. This presented no great problem in triggering the alarm, so long as the release occurred in a short time with a high tritium concentration. However, there was the possibility that the inverse would occur, that is, a low tritium concentration coupled with a long release time.

To correct these two deficiencies, it was decided to convert an ionization chamber output current proportionately to pulses and to feed the pulses to a counting system (Figure 2). We use a more sensitive version of an improved recycling integrator that was developed for current-to-frequency conversion of the photomultiplier tube output in the LLL automatic personnel dosimeter reader [2]. The seven-decade input current range (10^{-13} to 10^{-6} A) of this device lends itself well to the analog-to-digital conversion of the ionization chamber output current, thus, allowing integration of varying ionization chamber currents on a scaler. In the CTF (Current-to-Frequency) conversion technique, a MOSFET (Metal-Oxide-Semiconductor Field Effect Transitor) input converter is used (Figures 3 and 4). The MOSFET is a low-input-current solid-state device that functions as the input electrometer of the conductor. The converter yields a frequency (or pulse-rate) output directly proportional to input current at the rate of 10^{-11} A/Hz [3]. The CTF converter has good temperature-stability characteristics, drifting approximately 10^{-15} A/$^{\circ}$C at 25°C, and its 10^{-11} A/Hz transfer function is stable to better than 200 ppm/$^{\circ}$C above 10^{-11} A.

As finally designed (Figure 5), stack effluent from each of the two stacks is drawn through individual 1-liter ionization chambers at approximately 20 l/min (Figure 6). A potential of 1000 V is maintained

Figure 2

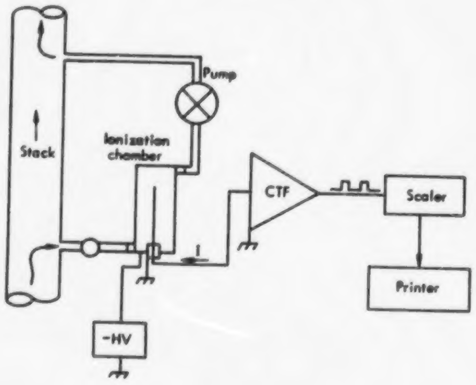


Figure 3

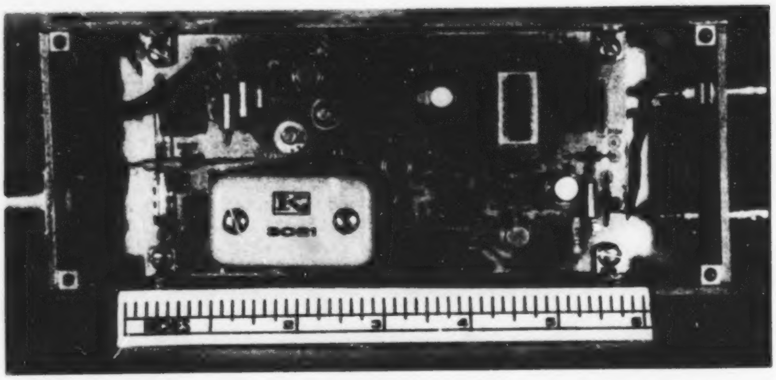
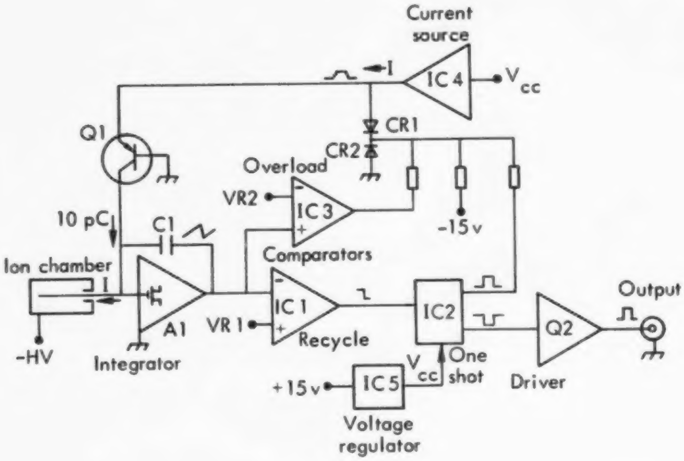
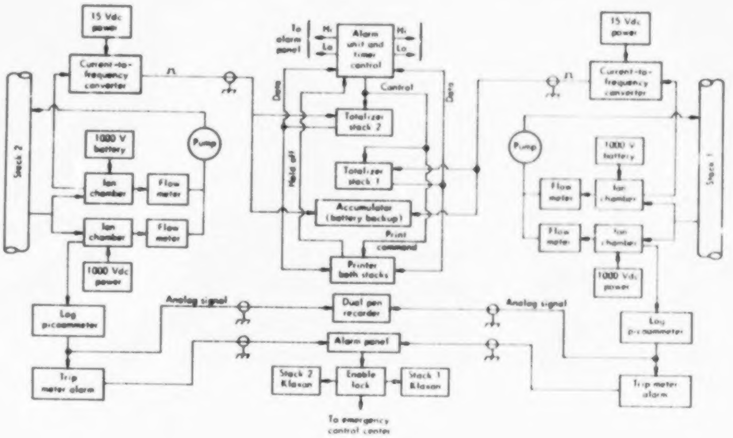


Figure 4



on each ionization chamber, to minimize nonlinearity due to recombination [4]. Each chamber output current is fed directly into a CTF MOSFET converter. Output pulses from each converter are counted by two parallel, seven-decade scalers located at the control console in the main office area (Figure 7). The converters are currently set to yield 10 counts/Ci.

Figure 5



One scaler, called the accumulator, is used for long-term recording of activity released from both stacks, and it may be reset manually only. Two buttons must be depressed simultaneously to reset this accumulator. One button is protected by a knurled knob that must be unscrewed before activating. These precautions were taken to eliminate the possibility of accidentally resetting the accumulator between monthly readouts. A special low-power scaler with COS/MOS (Complimentary-Symmetry/Metal-Oxide-Semiconductor) [5] integrated circuits was developed to permit use of low-power battery backup. This scaler normally operates on AC power and automatically switches to the battery without loss of accumulated counts in case of power loss, and requires only 12 Vdc at 100 mA (1.2 W) to operate.

The second scaler (one for each monitor) is a commercially available unit that also accumulates pulses from the CTF converter but is reset automatically every 15 minutes by an external timer. A 10-column printer prints the total count from the scaler before it resets on command from the timer. If the total count during any 15-minute period exceeds certain preset limits, a high- or low-level alarm is activated. The reset period and alarm points may be adjusted manually to allow for special circumstances.

The low-level alarm activates a bell within the building to warn occupants that a small release has occurred. If the high-level alarm is activated, the building klaxon sounds and a signal is sent to the LLL Emergency Control Center, which is manned 24 hours a day.

Figure 6a

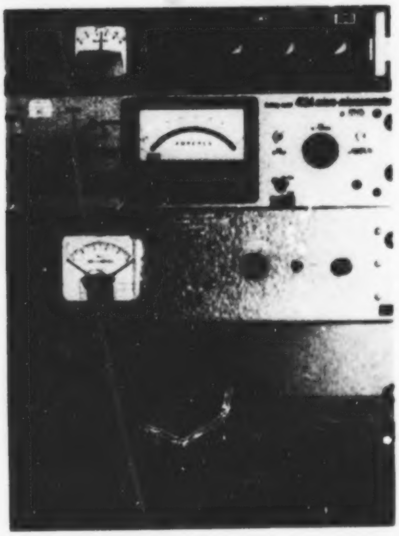


Figure 6b

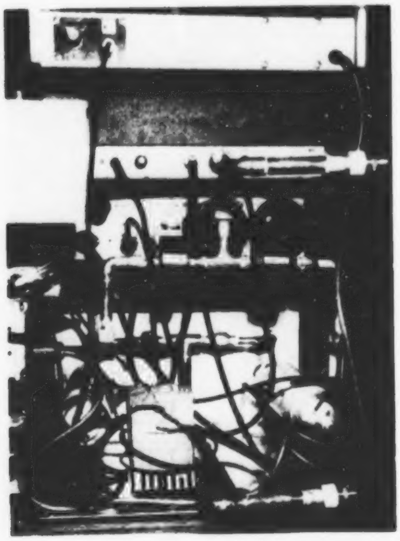
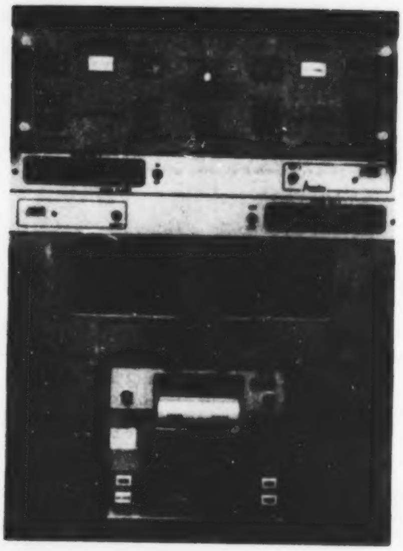


Figure 7



The stack integrator has been operational for about six months and has performed as accurately and reliably as originally planned. Total cost for the electronics, including installation, was under $5,000. The total system in its installed configuration has been calibrated at the lower end by comparing indicated release values obtained during known low-level releases. Upper-range response was checked by introducing known quantities of tritium to the ionization chamber and by exposing the chamber to a large radiation source. The original logarithmic picoammeter detection system will be kept in operation, since it provides valuable information about the release profile.

ACKNOWLEDGMENT

This report is based on work performed under the auspices of the U.S. Atomic Energy Commission.

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ON-LINE MONITOR FOR NATURAL GAS FROM NUCLEAR STIMULATION

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The Gasbuggy and Rulison experiments have established that tritium and krypton are the radionuclides of concern in nuclear stimulation of natural gas reservoirs. A system for continuous, on-line, simultaneous measurement of tritium and krypton radioactivity in natural gas has been constructed by Eberline Instrument Corporation for El Paso Natural Gas Company.

The on-line monitor system centers around an internal proportional counter. Feedback stabilization sensitive to pressure and chemical composition together with recorder read-out make the system capable of reliable, unattended operation for substantial periods of time. The system is built into a 30-foot trailer. The trailer has a self-contained living area and a lab area with a liquid scintillation counter and ancillary equipment.

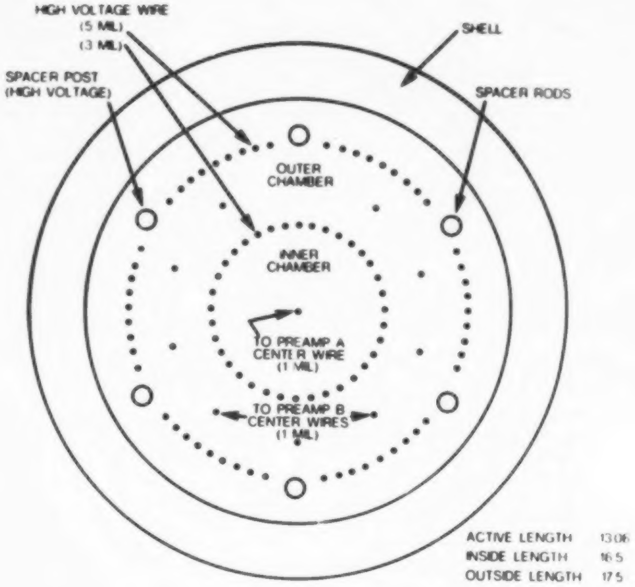
On-Line Monitor

The monitor system consists of a detector tube, stabilizer tube, high voltage supply, pulse height analysis electronics, scalers and printer, recorder, gas flow regulators and meters, and calibration gases.

The proportional detector has a grid structure as shown in Figure 1. Grid wires are strung on a framework to effectively form two annular chambers. Figure 1 is an end view of the grid wire arrangement. The active volume of the inner chamber, which measures tritium, is 670 ml of gas at STP or 1920 ml at the 42 psia operating pressure. The outer chamber is 5290 ml, while the total tube void volume is approximately 14 liters at operating pressure. Negative high voltage is applied to the grids while the center wires are used to develop the signal pulses. Gas flows through the tube parallel to the wires.

Figure 1

Detector Cross Section



The center chamber is designed so that low energy tritium betas emitted in this chamber have a small probability of penetrating the outer chamber. More energetic betas, such as those due to krypton-85, have a high probability of penetrating both chambers. The separation between tritium and krypton-85 is made on this basis as well as the difference in pulse height. The daughter products from any radon-222 which may be present in the gas will plate out primarily on the outer shell of the detector tube and not cause any appreciable buildup of background in the sensitive regions. The outer shell is a distance greater than the mean free path of an alpha particle from the sensitive region.

The stabilizer tube is about one-fourth the size of the detector tube. It contains a center wire surrounded by a plane of grid wires. A 1.4 μCi nickel-63 source (67 keV betas) is mounted adjacent to the chamber and produces about 10^6 cpm under normal operation. The count rate from the stabilizer tube is used to control the high voltage supply in a feedback loop. A block diagram of the loop is shown in Figure 2. The high voltage is about 4300 volts with pure methane in the tubes.

The operation of the stabilizer loop is the key to easy and accurate field measurement. Without this feature, minor changes in field gas would produce significant variation in pulse heights. If the gas gain (ion multiplication) in the stabilizer

Figure 2

Stabilizer Electronics Block Diagram

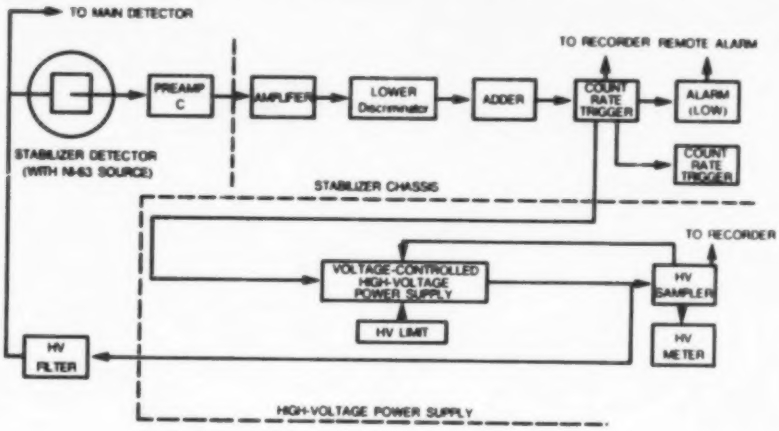
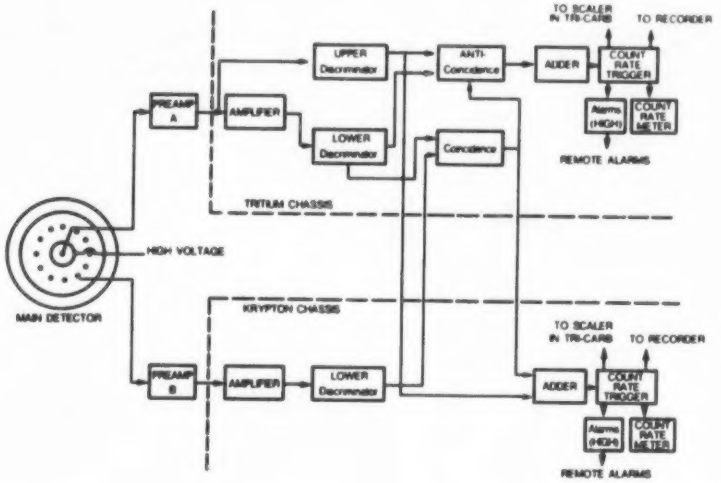


Figure 3

Detector Electronics Block Diagram



and detector tubes increases due to changes in temperature, pressure or chemical composition, the count rate of the stabilizer tube increases, causing the high voltage to be reduced. With this high gain feedback circuit, the pulse height is held essentially constant for wide variations of gas parameters. Since the stabilizer count rate is large compared to any count rate due to activity in the gas, the loop operation is essentially independent of gas activity level.

A block diagram of the electronics is shown in Figure 3. Preamp A is used for the center chamber while preamp B serves the outer chamber. Through level discrimination and coincidence gating, the upper channel counts pulses which are: 1) above preset noise level, 2) below 18 keV, and 3) generated only in the inner chamber. In other words, any pulse originating in the inner chamber is counted unless it is blocked in the anti-coincidence gate by a signal indicating that the pulse was also over 18 keV or signals from the coincidence circuits showing the pulse was seen simultaneously in both the inner and outer chambers. Thus, the upper channel counts essentially tritium. The lower channel counts pulses which are 1) above 18 keV or 2) above a preset noise level and coincident in both inner and outer chambers. Some of the krypton betas produce counts in the tritium channel, and the system is adjusted so that a few tritium betas are counted in the krypton channel. The concentration of each isotope is obtained through known calibration factors.

The counts from each channel are simultaneously registered by scalers and by count-rate meters with a logarithmic response. The output signal from each count-rate meter circuit is recorded by a multipoint recorder. The scalers are nominally part of the Packard Model 2311 Tri-Carb Liquid Scintillation Spectrometer. With this arrangement, automatic print-outs of data counted for periods between one second and 100 minutes are available for both channels.

The electronics section contains audio and visual alarms. If the count rates are outside preset values or if the gas gain cannot be maintained, an alarm is given. Relay contacts enable remote indicators to be used, thus permitting relatively unattended operation of this system.

A high voltage limit control is available to prevent excess high voltage when the gas quality is not suitable for proportional counting. Line frequency and voltage are metered and recorded.

A gas handling system is built into the trailer. It consists of piping, gauges, regulators, valves, and flow meters to supply gas from various sources to the tritium monitor. Flow meters and mixing chambers allow the dilution of field gas with pure methane if the field gas contains unusual constitutents or is highly radioactive. Bottled methane, tritiated methane, and a mixture of krypton-85 and methane are stored in a compartment at the rear of the trailer and are individually piped to the system. Simple manipulation of valves inside the trailer introduces each to the detector tube for background measurements or system calibration.

Operation

The on-line monitor is capable of operating at many different values of high voltage, pressure, discriminator levels, and amplifier gains. Consequently, the system can be adjusted for maximum performance under the conditions expected. The settings and operating points for which calibration factors are given are those believed best suited to measuring tritium at low levels, but with krypton-85 also present.

As previously mentioned, the detector high voltage is set to operate at about 4300 volts for pure methane. The discriminators are set so that some tritium is seen in the krypton channel. Typical background figures and calibration factors are shown in Table 1.

Table 1

Tritium Monitor - Background and Calibration Factors

Background:	Counts in tritium channel (B_T)	24 cpm
	Counts in krypton channel (B_K)	483 cpm
Calibration Factors:		
	Krypton in krypton channel (Kr_K)	6204 cpm/(pCi/ml)
	Krypton in tritium channel (Kr_T)	476 cpm/(pCi/ml)
	Tritium in tritium channel (T_T)	2834 cpm/(pCi/ml)
	Tritium in krypton channel (T_K)	34 cpm/(pCi/ml)

The system may be operated in a static or flowing mode. The flow rate used depends on the time constant desired for the measurement. Measurable flow rates as low as 100 ml/min or as high as 100 l/min may be used. Static measurement may be made on limited sample volumes.

Sensitivity

Comparative sensitivities can be calculated by assuming that the threshold for detection is two sigma above background. Choosing a reasonable counting time, the minimum detectable count rate may be calculated from $S = 2(B/t)^{1/2}$ where S is the signal count rate, B is the background count rate, and t is the counting interval. Multiplying S by a calibration factor gives the minimum detectable activity for one isotope in the absence of the other.

Table 2 compares the El Paso on-line monitor with a detector developed by the Bureau of Mines [1-3] and the LLL STALLKAT [4]. The Bureau of Mines' detector was originally used for tritium detection only. Another version of the detector was used at Project Rulison [5]. The LLL STALLKAT was the system used at Project Gasbuggy and at Rulison.

Table 2

Comparison of Tritium Monitors

	EPNG	BOM	STALLKAT
Tritium:			
Background (cpm)	24	720	50
Calibration factor (pCi/ml·cpm)	3.53×10^{-4}	1.6×10^{-4}	4.5
Detectable level (pCi/ml)	7.7×10^{-4}	1.9×10^{-3}	14
Krypton:			
Background (cpm)	483	720	400
Calibration factor (pCi/ml·cpm)	1.61×10^{-4}	1.1×10^{-4}	1.5×10^{-2}
Detectable level (pCi/ml)	1.6×10^{-3}	1.3×10^{-3}	0.13

However, such a comparison as this ignores the likelihood that both krypton-85 and tritium will be present, as well as the possibility of higher backgrounds due to Radon-222 which is likely to be in wellhead gas. If both krypton and tritium are to be measured, a more complex computation is advisable to determine the minimum counting time required to achieve a desired accuracy at particular levels of activity. If we arbitrarily choose to make one sigma equal to 10% of the count rate (a 69% chance of achieving 90% accuracy), and count background for a time equal to the signal count, the following relationship can be derived:

$$\text{for } {}^{3}\text{H},\ t = \frac{Kr_K^{\,2}\,(C_T + 2\,B_T) + Kr_T^{\,2}\,(C_K + 2B_K)}{0.01\,(Kr_K C_T - Kr_T C_K)^2}$$

where the symbols are explained in Table 1. A similar equation is used for krypton. Table 3 shows the results of several assumed situations for the backgrounds and calibration factors for the EPNG and BOM monitors.

Table 3 shows that reasonable count times may be used to accurately determine activity of 0.01 pCi/ml of one isotope with the other present and having an activity an order of magnitude greater. This assumes that the background count rate is stable over the total period for which signal and background are counted. The inevitable fluctuations in backbround make the minimization of background counts even more important. Measurements with the detector surrounded by bags of lead indicate that it would be possible to reduce the detector background to less than 5 cpm.

Table 3

Count Times to Achieve ±10% at One Sigma

Assumed Activity (pCi/ml)		Count Time Required (minutes)			
		EPNG		BOM	
Tritium	Krypton	Tritium	Krypton	Tritium	Krypton
0.01	0	10.2		49	
0	0.01		26.8		19
0.01	0.01	10.9	26.8	51	19
0.1	0.01	0.43	26.9	0.7	19
0.01	0.1	16.6	0.41	67	0.3

The upper limit of usefulness is determined by the dead time of the electronics, which has been measured at 3 μsec. At 600,000 cpm, a loss of counts of about 3% would have occurred. This corresponds to a krypton-85 concentration of almost 100 pCi/ml. The flow meters in the gas dilution system allow dilution ratios of 1:100 with an accuracy of 5%. Thus, concentrations as high as 10 nCi/ml may be measured with reasonable accuracy. Slightly higher activities of tritium may be reliably measured, and greater dilution factors are measureable, but with decreased accuracy.

The range of constituents which a gas may contain and still be within the counting limits of the system depends on the type of impurity and the dilution factors which may be used while retaining the desired sensitivity. Generally, pipeline and normal wellhead gases may be counted with only filtration and drying before introduction to the chamber. Methane, with up to about 20% by volume of carbon dioxide, nitrogen, or hydrogen, is easily handled by the system. Gas with larger percentage volumes of these contaminants may be counted by diluting with pure methane, but with corresponding loss of sensitivity. If significant volumes of air or oxygen (greater than about 2%) are included in the gas, dilution is necessary before counting.

The problem of additional background due to radon is a more serious one and less easily resolved. If the detector is used in conjunction with a tracer test, the background due to radon is simply added to any other background and the same equations hold. A stabilized flow of nontritiated gas containing radon can be introduced to establish background levels. In wellhead monitoring of a nuclear stimulation project, this is not possible since gas *identical* to that from the well is not available. Surrounding wells or predetonation gas may be used for comparison, but the radon-222 level may not be the same due to the gross changes in the reservoir caused by the nuclear explosion.

One method to surmount this difficulty is to obtain a simultaneous determination of the radon activity. Such a measurement is feasible with the EPNG monitor with a minor change to the electronics.

Results

The on-line monitor was used during a tracer experiment conducted in February 1971. The experiment was conducted jointly with Oak Ridge National Laboratory to determine the tritium distribution in plant gas and liquid products after large volume separation. This work was an extension of previous studies conducted jointly with Oak Ridge to determine the hypothetical dose to the general public through use of natural gas from nuclear stimulation [6].

Natural gas from the Gasbuggy nuclear stimulation experiment near Farmington, NM, was transported to a gas processing plant in the Permian Basin. The tracer gas, having a tritium concentration of about 12 pCi/ml, was blended with input field gas to the plant, which was decoupled from the El Paso system. The on-line monitor sampled the plant output gas prior to flaring. Under steady state flow, the measured tritium activity was about 0.3 pCi/ml and the krypton-85 was about 0.06 pCi/ml. The background level in the tritium channel was 29 cpm. At the conclusion of the test, the on-line monitor indicated radioactivity levels less than 0.002 pCi/ml for both isotopes. After tracer cutoff, the combined time constant for the plant and monitor was about 25 minutes.

Conclusions

The Eberline design, utilizing an automatic stabilization system, is an evolution from the Bureau of Mines' design which incorporates a system for manual calibration. The use of annular geometry for the detector has two advantages over the use of four square chambers as incorporated in the Bureau of Mines' detector. Background counts in the tritium channel are reduced by a factor of 20 to 30 and the rejection of high energy betas as in krypton-85 is increased by about a factor of 10.

The on-line monitor provides a fast and accurate method for measuring tritium and krypton-85 activities in natural gas. The system is versatile enough so that extensive analyses and calibration procedures are not necessary prior to use. The trailer mounting allows operation in a wide variety of locations and allows relatively easy movement.

The on-line monitor can measure concentrations of tritium and krypton-85 as low as 10^{-2} pCi/ml and as high as 10 nCi/ml. The system operates automatically and offers reliability of measurement by adjusting for and providing indirect evidence of compositional or other changes in the gas measured.

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CHAPTER EIGHT
Environmental Monitoring Data

TRITIUM RELEASES FROM NUCLEAR POWER STATIONS

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Tritium in effluents from nuclear power stations and associated facilities is of particular interest because it is the most prevalent radionuclide in liquid wastes [1-3] and is exceeded among long-lived radionuclides in gaseous effluents only by ^{85}Kr. Fortunately, the radiation hazard per curie of tritium is low compared to most other radionuclides, because tritium releases relatively little energy per disintegration and has a short residence time in the human body. Nevertheless, due to its relatively high concentrations, tritium may be the critical radionuclide for calculating population radiation exposures at a number of nuclear power facilities.

Detailed discussions of sources of tritium and its behavior in the environment have recently been published [4-7]. Data on tritium discharges are reported periodically by nuclear power plants, and are summarized for or by the Atomic Energy Commission (AEC) and the Environmental Protection Agency (EPA) [1-3]. As indicated below, however, there is still considerable uncertainty concerning tritium production and discharge. Important aspects of tritium behavior have been obscured by the difficulty in detecting tritium other than by specific analysis for the radionuclide. For example, its production by ternary fission, the main source in nuclear power reactors, was not observed until 1959, and the AEC did not require routine reports of tritium releases by nuclear power stations until 1965.

In this paper, the release of tritium to the environment at routinely operating nuclear power stations and at a fuel reprocessing plant is reviewed on the basis of production estimates, operating reports by these facilities, and data from radiological surveillance studies. The latter were performed at a boiling water reactor (BWR), a pressurized water reactor (PWR), and a fuel reprocessing plant. Some of the other contributors to environmental tritium, experimental and test reactors, research laboratories, tritium production plants, nuclear explosions for weapon testing and peaceful applications, and industrial and medical users, are considered elsewhere in this book.

Tritium Production

The obvious sources of tritium in the water, boron control rods or solution, and uranium fuel of power reactors, are listed in Table 1 on the basis of values suggested by the General Electric Company and Westinghouse Corp. [8,9], the respective suppliers of most U.S. BWR and PWR systems. Tritium is also produced in reactions of neutrons with lithium, especially ^{6}Li (as trace impurity or product of a reaction between neutrons and boron), ^{14}N (in air, cover gas, or ammonia added

Table 1

Estimated Tritium Production at Nuclear Power Stations, Ci/a (Power Level Approximately 1000 MWe and 3300 MWt)

Source	Reaction	BWR [8]	PWR [9]
coolant water	$^{2}H(n,\gamma)^{3}H$	8	3
dissolved boron	$^{10}B(n,2\alpha)^{3}H$	0	600
uranium fuel rods	fission	13,000	13,000
boron control rods	$^{10}B(n,2\alpha)^{3}H$ and $^{10}B(n,\alpha)^{7}Li$, $^{7}Li(n,n\alpha)^{3}H$	5,000	0

to the water), ^{3}He (formed in the decay of ^{3}H), and ^{239}Pu (formed in fuel by neutron irradiation of uranium) [4,5,9-12]. The reported production rates, discussed in detail in several papers of this book [13-16] are generally consistent with observed tritium concentrations in coolant water and amounts of tritium in liquid wastes. Confirmation of computed values by such means is quite approximate in view of the many sources of tritium, the changing amounts of these sources during reactor operation, and the complexity in defining the distribution of neutrons and sources within the reactor.

Discharges to the environment at nuclear power stations include the tritium formed in water (to a first approximation, the amounts in the first two lines of Table 1) plus the fraction of the tritium formed in fuel and control rods that enters the water. At Dresden, a BWR station with zircaloy-clad fuel, concentrations in the coolant water suggest that considerably less than one percent of the tritium was diffused through cladding into the cooling water [8]; at Ginna, a PWR station that uses zircaloy-clad fuel, approximately one percent of fission-produced tritium was found in the primary water [9]; at four other PWR stations (built earlier and using stainless steel cladding), the amounts in the coolant water and liquid wastes are more consistent with tritium leakage rates of either 16% [12] or 50% [17]. Some of the measurements also suggest that the instantaneous fractional transfer from the fuel deviates from long-term averages, possibly as a result of local temperatures, cladding imperfections, or accumulation of gas within the rods.

Thus, if coolant water is discharged as frequently as is now the case, the tritium passing to the environment at nuclear power stations may be approximately one percent of total generation if zircaloy-2 cladding is used, or 50% with stainless steel cladding. The residue remains in the used control rods that are shipped off-site for solid waste disposal, and in used fuel rods that are transported to the reprocessing plant. Changeover to zircaloy-clad fuel at the four PWR stations that now use stainless steel cladding should reduce considerably the amounts of tritium discharged to the environment, and increase the tritium retained in fuel elements. Further reduction of tritium discharges by increased recycling of system water is being contemplated [9]; on the other hand, as nuclear power stations become larger and more numerous, total discharges will undoubtedly increase.

At the fuel reprocessing plant, the tritium in the fuel may be discharged with waste liquids and airborne effluents, or may remain with the solid wastes (undissolved cladding material) and high-level liquid wastes. Most of the tritium in the dissolved portion is believed to be discharged with liquid wastes in the form of water, although some gaseous waste in the form of water and elemental hydrogen has been reported in pilot plant studies [4]. It is uncertain whether the fraction of tritium in the stack effluent is 1, 20, or 25 percent of the total [4].

Tritium Discharges

The record to date of tritium discharged to the environment in liquid effluents at full-scale commercial U.S. nuclear power stations, Table 2, shows a few curies per year at each BWR station and hundreds or thousands of curies per year at each PWR station. These amounts are qualitatively consistent with the sources described above. The variations in discharges at each station can be explained only in part by variations in power level, as occasioned, for example, by initial operation at low power or reactor shut-down for part of the year. More data would be required to predict annual releases at large BWR and PWR stations that use zircaloy-clad fuel.

Limited information on tritium in airborne effluents suggests that these are a minor path to the environment. Yankee operating reports give releases between 8 and 16 Ci/a in 1965-1970, Connecticut Yankee, between 0.1 and 9 Ci/a in 1968-1970, and San Onofre, 11 Ci/a in 1970; two measurements at Dresden suggest a discharge of approximately 0.2 Ci/a [18]. All the cited values are for tritium in the form of water. Several measurements at Yankee indicate that some of

Table 2

Tritium Discharge in Liquid Waste,* Ci/a

Plant	Initial Operation	Power, MWe	System	1965	1966	1967	1968	1969	1970
			Power Generation						
Dresden I	1959	210	BWR	5-10**	5-10**	( )**	3	~6	5
Yankee	1960	185	PWR	1,300	1,920	1,590	1,170	1,200	1,490
Indian Point	1962	265	PWR	490	120	300	790	1,100	410***
San Onofre	1967	450	BWR	---	---	---	2,350***	3,500	4,780
Conn. Yankee	1967	600	PWR	---	---	120***	1,760	5,200	7,380
Oyster Creek	1969	515	BWR	---	---	---	---	5***	22
9-Mile Point	1969	500	BWR	---	---	---	---	---	20***
Ginna	1969	420	PWR	---	---	---	---	---	107***
			Fuel Reprocessing						
Nuclear Fuel Services	1966	---	---	---	290***	4,200	2,610	5,980	4,520

*Data for Nuclear Fuel Services, 1966-9, reference 2; Indian Point, 1965, and Dresden I, 1965-6, reference 18; other data for nuclear power stations, 1965-6, reference 3; and for 1967-9, reference 1; all plants, 1970, reference 19.

**Estimated; no value found for 1967.

***Not operating full year.

the effluent tritium gas is not water vapor, but possibly elemental hydrogen or some other gaseous compound [19].

At the fuel reprocessing plant, approximately 5 kCi/a were discharged in liquid waste during 1967-1970, as shown in Table 1. Two measurements during fuel dissolution in 1969 of tritium as water vapor in stack effluent indicated 1.0 Ci ^3H per 10 kCi ^{85}Kr [20]; at ^{85}Kr discharges between 180 and 328 kCi/a during 1967-1970 [2,21]; tritium discharges to air would have been between 18 and 33 Ci/a. Recent unpublished measurements by Cochran suggest that additional tritium, released continuously during fuel processing, may have increased the total airborne release of tritium to approximately 100 Ci/a. Tritium could also be detected as a gas (not water vapor), in small, unquantified fractions of the total airborne tritium [20].

Tritium in the Environment

The highest annual average concentrations in air and water at the boundaries of the nuclear power facilities, computed from the data in the preceding section, are shown in Table 3 to be very small fractions of the AEC concentration limits [22]. The indicated concentration in water is readily detectable by counting samples directly with liquid scintillation counters, whose limits of detectability are

Table 3

Highest Annual Average Tritium Concentrations in Air and Water at Plant Boundary

	Air	Water
Location	Yankee	Conn. Yankee
Year	1965	1970
Annual discharge, µCi	1.6×10^7	7.4×10^9
Dilution medium	ventilating air	circulating coolant water
Dilution medium flow rate, ml/min	4.2×10^8	1.4×10^9
Annual dilution volume, ml	2.2×10^{14}	7.4×10^{14}
Further dilution factor to boundary	1.0×10^3	1.0
Annual average concentration at boundary, µCi/ml	7.3×10^{-11}	1.0×10^{-5}
Annual average concentration limit, µCi/ml	2.0×10^{-7}	3.0×10^{-3}
Fraction of limit	4.0×10^{-4}	3.0×10^{-3}

Table 4

Tritium in Environmental Samples at Nuclear Power Plants

Plant	Sample	Highest Tritium Concentration, pCi/ml
Yankee, 1969	Circulating coolant outlet water during liquid waste discharge [17]	220
	Yard drain effluent water [19]	79
Indian Point, [19] 1969	Circulating coolant outlet water during liquid waste discharge	15
Conn. Yankee, [24] 1970	Circulating coolant outlet water during discharge of liquid waste	65
Nuclear Fuel Services, 1966-7	Cattaraugus Creek water during liquid waste discharge [25]	185
	Cattaraugus Creek fish [25]	1
	Air [23]	11
	Deer (soft tissue) within exclusion fence [25]	46

usually between 0.1 and 1 pCi/ml. The tritium in air as water vapor is also detectable if the vapor is collected by condensation or absorption [23]: at a water content of 2 ml/m^3 of air, the tritium concentration in air would be 36 pCi/ml of water. Annual average concentrations of tritium at the boundaries of the two stations during other years, and at all other plants, were lower; at the BWR stations, they would have been undetectably low because of the far lower discharges and higher atmospheric dilution factors.

The wastes that contain the highest tritium concentrations at PWR stations and the fuel reprocessing plant are usually discharged intermittently, hence environmental samples would be expected to show no detectable tritium on some occasions, and concentrations higher than annual averages on others. The highest concentrations of tritium in environmental media given in facility reports and surveillance studies are listed in Table 4. Average values were considerably lower. All concentrations are below the AEC limits for water and air at the boundary; of the samples, only the fish at Nuclear Fuel Services might have been ingested by humans. These detectable levels in analyzed samples and the above calculations suggest that initiation of tritium analyses of water, fish, and air should be considered during discharges of high-tritium wastes by PWR stations that have not reported such measurements.

The contribution by these nuclear power facilities to the worldwide amount of tritium has, so far, been trivial, as indicated by the discussion of environmental tritium in other papers in this book [26]. In brief, the total discharge in 1970 of 20 kCi, Table 1, compares with the estimated amounts of 20 to 200 MCi of naturally produced tritium and 1000 MCi tritium from atmospheric weapon testing at this time [4,5]. These values may be referred to concentrations in terms of 1.6×10^{22} g hydrogen available for dilution, mostly in oceans (i.e., 1.4×10^{23} ml water), and 4.3×10^{24} ml atmospheric volume [27]. At a thousandfold increase in production of nuclear power, and thus, of tritium, during the next three decades [4], the worldwide steady-state levels (18 times the annual discharge rate) of 300 MCi would exceed the natural background but still be below present amounts.

Summary and Conclusions

Discharges of tritium to the environment at full-scale commercial U.S. nuclear power stations and one fuel reprocessing plant have been steadily increasing and amounted to 19 kCi in 1970. Almost all of the tritium was discharged into water, but a small fraction of the effluent tritium was discharged through the stack as water vapor and gas. Most of the 19 kCi was released at pressurized water reactors and the fuel reprocessing plant. At the PWR stations, the tritium is believed to have been produced mainly by ternary fission and to have diffused through stainless steel fuel cladding into primary coolant water, which was later discharged. The fission-produced tritium that remained in the fuel was released at the reprocessing plant during fuel dissolution.

Tritium would be expected to be readily detectable while some wastes are being discharged, at PWR stations and the fuel reprocessing plant, into air, water, and media whose water content rapidly reaches equilibrium with the air and water. Tritium has been found in some of these samples, at concentrations well below AEC limits. Tritium concentrations in environmental samples at BWR stations would not be expected to be noticeably above background levels. The contribution of discharged tritium to worldwide levels is, at this time, negligible and undetectable.

Further research is needed in several areas. Production rates of tritium by some of the many reaction mechanisms possible in power reactors are uncertain, as are the transfer rates of tritium from fuel to coolant water under the influence of various operating variables. Further information is required on the amounts and chemical forms of tritium discharged to the air at nuclear power stations and fuel reprocessing plants. To supplement the inferences from discharge data on which exposure calculations are often based, inclusion of sampling and analysis for tritium is needed in environmental surveillance programs to provide measured values for computing population radiation exposures.

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ENVIRONMENTAL TRITIUM CONTAMINATION FROM NUCLEAR POWER PROGRAM

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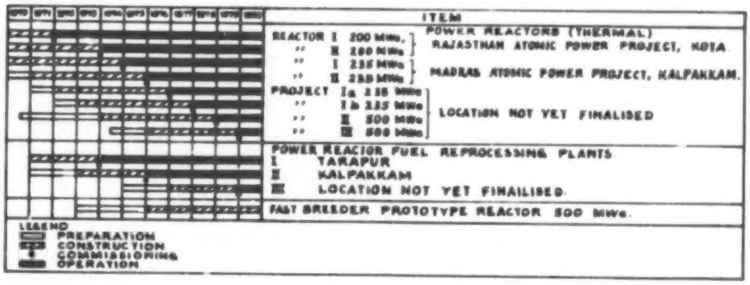
The estimate for the growth of electrical power in India by the end of the fourth five-year plan, i.e., 1974, is 23 GWe. Assuming a consumption growth rate of 12%, the country will need about 45 GWe of installed capacity by 1980. The Indian Atomic Energy Commission, in its recent publication on the growth of atomic energy, has proposed that about 2700 MWe power should be generated by 1980 from nuclear energy [1]. The Commission has indicated that these nuclear power stations will be of CANDU design, utilizing heavy water as both moderator and coolant and natural UO_2 as fuel, Figure 1. By 1980, three fuel reprocessing plants are expected to process the irradiated fuel from the power reactors. It is proposed that one fast breeder reactor (40 MW) will be built to provide a base for the fast breeder power stations expected after 1980.

Tritium is a radionuclide of interest because of its radiological properties and appreciable production. It is released to the environment as a consequence of normal operation in both nuclear power reactors and fuel reprocessing plants.

Figure 1

Milestone Chart for Atomic Power Projects and Fuel Reprocessing Plants



Tritium may be produced in nuclear reactors by ternary fission of the isotope of ^{235}U, ^{218}U, and ^{239}Pu having triton as one of the fission fragments, or by activation reactions with coolant, cladding and other materials according to the following reactions:

$$D_2O\ (n,\gamma)\ TDO$$

$$H_2O\ (n,\gamma)\ DHO\ (n,\gamma)\ TDO$$

$$^{10}B\ (n,\alpha 2)\ T$$

$$^{10}B\ (n,\alpha)\ ^{7}Li(n,\ n\alpha)T$$

$$^{6}Li\ (n,\alpha)T$$

$$^{11}B\ (n,\ n2\alpha)T$$

The relative importance of a given reaction mechanism for tritium production will depend upon the reactor type, design characteristics, operating history and materials of construction. Depending on these factors, the contribution to tritium production from one of the main reactions mentioned may be large compared to others.

Tritium Production in Nuclear Fission

Tritium as one of the products of nuclear fission was first reported by Albenesius [2]. He established that tritium is produced

Figure 2

Fission Product Tritium Production in Pu and U Fuel

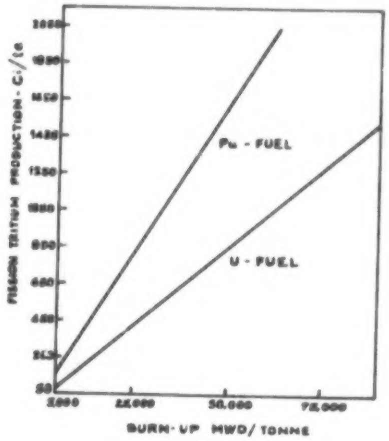
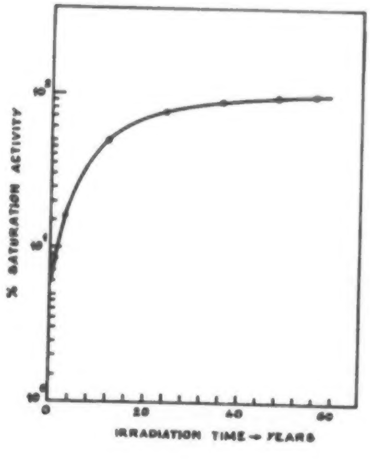


Figure 3

Build-up of Tritium Activity in Heavy Water with Irradiation Time



at the rate of one atom per 1 to 2 X 10^4 thermal fissions of uranium. Theoretical fission yields have since been calculated for fissionable materials (uranium and plutonium), for both thermal and fast fissions. Measurements of the tritium released in fuel reprocessing indicate that the theoretical estimate of 1.3 X 10^{-4} tritium atoms/fission is the most accurate [3]. Fast reactors using ^{239}Pu as fuel produce more fission product tritium than thermal reactors, since the tritium yield in fast fission of ^{239}Pu is higher than in thermal fission of ^{235}U (2.5 X 10^{-4} and 1.3 X 10^{-4} atoms/fission, respectively). Using these values, the production of fission product tritium in uranium and plutonium fueled reactors for various burnups is given in Figure 2. The tritium production rates for U and Pu fuel were calculated to be 17 mCi/MW-d and 32.6 mCi/MW-d, respectively. These values do not take into account the residence time for the fuel and the load factor for the reactor. Fuel cladding will, however, retain the tritium since very little diffusion has been reported through stainless steel or zircaloy cladding.

Tritium Production in Activation Reactions

Light-water reactors

Tritium can be produced by neutron reactions with different elements, the major ones being the deuterium and ^{6}Li in the coolant system [4]. For the Tarapur Atomic Power Station (BWR type, Power 660.9 MWt), tritium production due to the deuterium activation is calculated to be at the rate of 2.2 nCi/ml per month and an equal contribution from traces of lithium. Tritium measurements until April 1971 in reactor coolant samples show concentrations ranging between 0.38 and 1.7 nCi/ml. This lower value could be due to losses and the make-up of the coolant. This order of tritium contamination is so low in comparison to the other activation and fission products in BWR stations that it has not received much attention.

Heavy-water reactors

This system uses 99.76% pure heavy water as both coolant (a high temperature and high pressure system) and moderator (a low temperature and low pressure system). The build-up of tritium activity with irradiation is given in Figure 3. The amount of tritium produced in the moderator is about fifty times higher than that produced in the primary heat transport system, as is evident from Table 1. Being a low temperature, low pressure system, the leakage of heavy water from the moderator is not likely to contribute significantly to the leakage of tritium from the station, assuming that there is no mix-up of D_2O from the moderator and Primary Heat Transport (PHT) systems. Holmquist [5] suggests that leakages of heavy water could realistically average 2 to 3% of the annual inventory. The total tritium activity contained in the moderator and PHT systems of the proposed power reactors built in this decade is predicted to be nearly 57 MCi by the year 2000.

Table 1

Saturation Tritium Activity in PHT and Moderator System for Reactors Planned for Period 1971-80

Power Reactors*	Power MWe	Start-up by	Saturation activity (MCi)	
			PHT System	Mod. System
RAPP I	200	1972	0.117	6.4
RAPP II	200	1974	0.117	6.4
MAPP I	235	1975	0.120	7.5
MAPP II	235	1976	0.120	7.5
Project Ia	235	1977	0.120	7.5
Project Ib	235	1978	0.120	7.5
Project II	500	1979	0.254	16.0
Project III	500	1980	0.254	16.0

*For details of location, etc., see Figure 1.

Tritium Releases From Nuclear Reactors

Fission product tritium

The bulk of this tritium is retained in the fuel and is not released until the fuel elements are processed in fuel reprocessing plants or get damaged during the operation of the reactor. The type of fuel cladding has been reported [6] to have an appreciable effect on the amount of tritium retained in the fuel. Tritium measurements in the coolant of BWRs indicate that the tritium release rate is higher in stainless steel clad fuel than zircaloy clad fuel. In CANDU reactors, the contribution due to fission product tritium released from the failed fuel is extremely small as compared to the tritium production in the heavy water.

Activation product tritium

Releases of tritium from light boiling water reactors are to be insignificant compared to other activities. The reported [7] yearly release from Dresden I is only 5 to 10 Ci. Measurements at Tarapur show a release to the coastal sea waters of about 10 Ci/a through liquid wastes. The operation of the research reactor CIRUS at Trombay gives rise to stack discharge of tritium, which, based on the present tritium concentration in the heavy water, is measured to be about 1.5 Ci/day. This results in measurable air concentration values at various locations on site (ranging up to 170 pCi/l of air). Though not of radiological consequence, these measurements have led to a detailed study [8] of dilution rate factors for the reactor stack.

The release from CANDU reactors is estimated to be much higher. It has been reported [9] that D_2O leakage from the Douglas Point Generating Station (DPGS) in Canada is of the order of 4 kg/d, i.e., 0.6% annually. Wilson et al. [10] indicate that about 90% of the total tritium released from Douglas Point goes to the environment via the stack, and 10% is discharged to the liquid effluents. Typical figures for these releases are about 1,000 Ci/month through the stack and 100 Ci/month to the liquid effluents, with a PHT tritium activity of 1.6 to 1.9 Ci/l. On the basis of this experience at Douglas Point, tritium released from RAPP I and II is expected to be about 50 KCi/a through stack discharge and about 5 KCi/a through liquid effluents to Rana Pratap Sagar (RPS) dam at saturation tritium activity. Similar situations are expected for MAPP I and II, Project I (a and b), Project II and Project III.

Tritium Release From Fuel Reprocessing Plants

In fuel reprocessing plants, nearly all of the fission product tritium is available for release to the environment. It is estimated that in the Purex process, about 10% of tritium activity is retained with the high level wastes, about 65% is disposed to surface water through low level effluents and about 25% is discharged to the atmosphere through the stack. Measurements at the Trombay reprocessing plant indicate that tritium release to low level wastes amounts to less than 100 mCi/d. Stack release does not contribute significantly compared to CIRUS stack release.

The PREFRE plant (capacity 0.5 te/d) at Tarapur will process fuel from the Tarapur Atomic Power Station and Rajasthan Atomic Power Station. The expected tritium releases from this plant are given in Table 2 for both cases, the processing of TAPS fuel and RAPS fuel.

Table 2

Estimated Tritium Release from PREFRE

Estimated Burn-up	Tritium Content of Fuel Ci/ton	Estimated Average Release Rates (Ci/d)		
		Stack 25%	Low Level Liquid Waste 65%	High Level Active Waste 10%
12,000 (TAPS)	200	25	65	10
6,500 (RAPS)	110	14.75	34.75	5.5

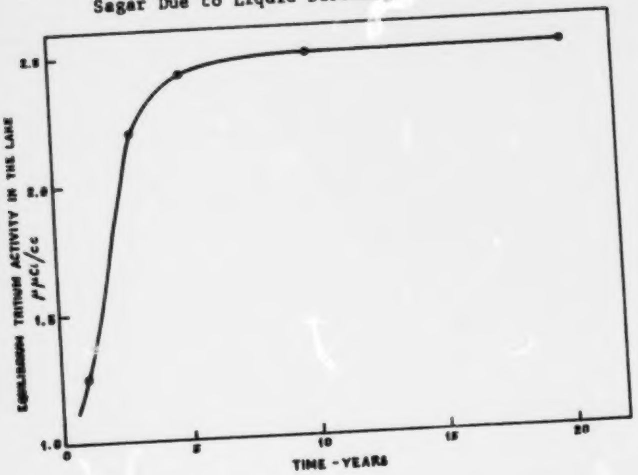
For the processing plant at Kalpakkam (Madras) the values given in Table 2 for RAPS fuel will be applicable, with about 10% extra contribution from processing of fuel from FBTR (40 MWt) fast reactor.

Tritium in RAPP Environment

Tritium, regardless of its mode of production or release, eventually deposits in the hydrosphere. The highest environmental concentration of tritium would be expected to exist in the area surrounding the point of release.

Figure 4

Build-up of Tritium Activity in Ranapratap Sagar Due to Liquid Discharges from Raps



Release to surface waters

For RAPS, Rana Pratap Sagar is the source of water for the condenser cooling system. This lake has a volume of about 2.83 X 10^9 m^3, and the flow of water through it varies from 50 to 230 m^3/sec. Normal operation of RAPS I and II is expected to lead to an average tritium release of about 5 KCi/a into the lake. Tritium concentration in the lake can be calculated using the model proposed by Peterson et al. [11]; build-up of tritium activity is given in Figure 4. Saturation tritium activity in the lake on the basis of lowest flow works out to 2.5 pCi/ml, which is about 1,000 times less than the MPC_w for members of the public.

Release to atmosphere through stack

In the case of stack release, Cowser et al. [12] consider the effect of washout and deposition on clouds and conclude that the ground level air concentrations would not be reduced by washout. Land surface can act either as a source or a sink, depending on the magnitude and direction of water movement.

Studies of the micrometeorological parameters at RAPP over years indicate that an average dilution factor of about 10^{-8} sec/m^3 is obtained at 1 km from the station for discharge through a 100-m stack. With an expected discharge rate of about 50 kCi/a, environmental ground level concentrations are expected to be about 0.1 pCi/m^3, compared to MPC_a of 0.2 μCi/m^3 for individual members of the public.

A similar situation is expected from stack releases at Kalpakkam from MAPP I and II, with a 10% extra contribution from fuel reprocessing plant discharges. As this is a coastal site, the liquid discharges will be to nearby coastal waters through a condenser cooling system, and will thus not lead to contamination of surface waters.

Conclusion

An attempt has been made to estimate the sources of tritium available for release to the environment as a result of the nuclear power program during this decade (1970-1980). Environmental consequences at two sites, RAPP and Kalpakkam, are shown to be insignificant from radiological safety considerations under normal conditions.

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TRITIUM SOURCES AT THE SOUTHWEST EXPERIMENTAL FAST OXIDE REACTOR

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The production of tritium at nuclear facilities during the past decade has received considerable attention [1,2]. Tritium levels have been observed and reported for light water and heavy water cooled reactors [3,4]. More recently, studies at the EBR-II sodium-cooled reactor have shown tritium to be present throughout both the primary and the secondary heat transfer systems [5,6], although the levels are generally lower than in water reactors.

The presence of tritium at the Southwest Experimental Fast Oxide Reactor (SEFOR) Facility in minute, but measurable, quantities has led to an examination of tritium sources at SEFOR. The use of nitrogen gas, which blankets the primary system and also acts as a coolant for control reflectors and a biological shield located in the high neutron flux immediately surrounding the reactor pressure vessel, constitutes a tritium source unique to SEFOR [7]. A description of this and other sources of tritium and a discussion of the significance of each are presented in this paper.

To understand the tritium production sources at SEFOR requires some familiarity with the salient features of the facility. The reactor is a 20 MW (steady-state), fast, sodium-cooled reactor with mixed PuO_2-UO_2 fuel. SEFOR was designed to measure the physics characteristic of a mixed oxide fueled, sodium-cooled fast breeder reactor, particularly the Doppler Coefficient which is a prompt-acting inherent control feature. The experimental program also includes series of tests to demonstrate the effectiveness of the Doppler to limit the reactor power resulting from rapid insertions of large amounts of reactivity. Neutron spectrum and fuel temperatures in SEFOR were established to be typical of those expected in a large sodium-cooled breeder reactor used for commercial electrical power production.

Argon is used as the cover gas over the sodium in the vessel. The refueling cell surrounding the reactor head also contains argon during reactor operation. The argon is normally maintained at a very high level of purity with respect to oxygen and moisture constituents, i.e., <10 ppm O_2 and moisture.

Nitrogen gas is used in the zone surrounding the primary sodium heat-exchanger loop, and also cools the reflectors and guide structures which constitute the reactor control mechanism. The nitrogen zone may contain a few percent of oxygen.

Tritium in Sodium

Measurements of sodium samples, obtained quarterly, have shown a small but measurable quantity of tritium. This component does not constitute a serious health physics problem because of its low level and because the radiological, toxicological and pyrophoric characteristics of the sodium already require certain precautions. It is of interest, however, to evaluate the methods by which tritium can be introduced into the sodium, and to compare the measured quantity present with that predicted.

The three methods by which tritium may be produced in the primary sodium are ternary fission, boron activation and lithium activation. The production of tritium as a result of ternary fission in nuclear reactors has been extensively described. Based on the value for the thermal fission yield of tritium (one tritium atom per 1.25×10^4 fissions)[2] calculations indicate that 1.8×10^{17} atoms of tritium are produced in the SEFOR reactor for one Equivalent Full-Power Hour of Operation (EFPH). Recent information indicates that a higher yield may be expected for fast neutron fission. This corresponds to a production rate of approximately 8.74 mCi per EFPH.

Tritium from ternary fission is produced in both fuel rods and tramp uranium in the system. The fuel consists of small ceramic pellets contained in a stainless steel cladding. The thickness of the cladding is 40 mils, which would tend to mitigate against recoil penetration. However, at the core midplane, the estimated fuel temperature is approximately 4500°F, so the diffusion process through the fuel would be enhanced. The ternary fission process in the fuel appears to be a significant contributor to tritium in the primary sodium. From other data, the extent of tramp uranium has been estimated to be of the order of milligrams, making the contribution from this source negligible.

Trace quantities of boron have been measured in the SEFOR primary sodium at levels of approximately 2 ppm. This corresponds to a ^{10}B content of approximately 4.8×10^{23} atoms in the primary system. If a cross section of 25 mb is assumed for the $^{10}B\ (n,t)\ 2\alpha$ reaction and an effective neutron flux above the reaction threshold of the order of 1×10^7, then 2.4×10^4 atoms of tritium are produced per second at 20 MW or 8.6×10^7 atoms per EFPH. This source is insignificant compared with that produced from the ternary fission process shown above.

Tritium may also be produced from the $Li(n,n\alpha)t$ reaction which has a 50 mb cross section for neutron energies above 4 MeV. Calculations based upon an examination of trace materials in sodium indicate a lithium-7 content of approximately 4.5×10^{24} atoms. This amount would yield 2.3×10^6 atoms of tritium per second at 20 MW, or 8.3×10^9 atoms per EFPH. Again, the production of tritium from this source appears to be insignificant as compared with the fission yield.

Tritium in the Nitrogen Zone

The presence of a large volume of nitrogen gas in conjunction with a fast neutron flux provides a mechanism for the production of tritium at SEFOR which is somewhat unique. Tritium is produced in the nitrogen zone by the following reactions:

$$^{14}N\ (n,t)\ ^{12}C$$

This reaction has an effective neutron energy threshold at about 3.7 MeV, and an effective cross section of 0.01 barns, as averaged over the fission spectrum. Calculations [7] indicate a production of 3.0 X 10^{10} atoms per second for a reactor power of 20 MW.

Tritium is also produced by the irradiation of boron contained as B_4C on the serpentine rods. The cross section for the reaction, ^{10}B (n,t) 2α is 0.075 barns for a neutron energy above 3.7 MeV, and 0.025 MeV for a neutron energy from 1.3 to 3.7 MeV. These data provide a calculated tritium yield of 1 X 10^{10} atoms per second from the boron source [7], if essentially all of the tritium is released to the nitrogen.

Comparison of Calculated and Measured Tritium Levels in SEFOR in the Primary Sodium

Because of the relatively long half-life of tritium, production from each of these sources is essentially a time dependent function of reactor operation and power. Between December 1970 and February 1971, the reactor was engaged in a power ascension phase of the program, with a consequent rapid increase in the integrated reactor power. Figure 1 presents a curve of the integrated power from December, 1970 through June, 1971.

Figure 1

Reactor Operating History

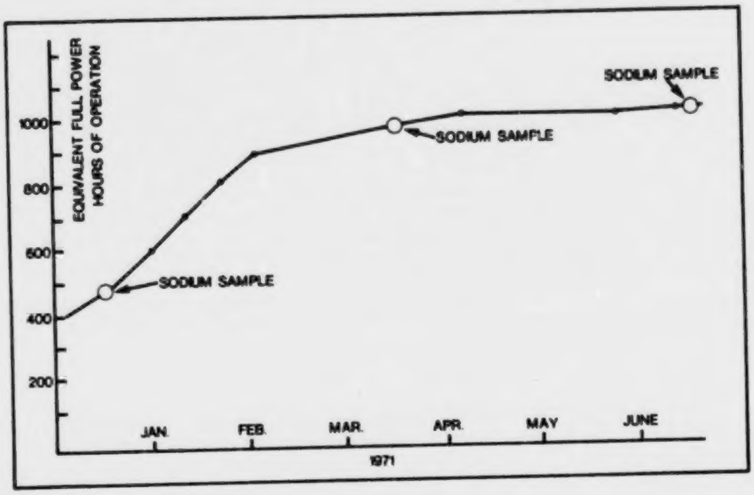


Table 1

Tritium Content of SEFOR Primary Sodium

Date	EFPH	Tritium Concentration in Primary Sodium (μCi/g) Calculated	Measured
12/17/70	480	0.41 ± 0.04	0.28 ± 0.20
3/16/71	970	0.83 ± 0.09	0.87 ± 0.04
6/16/71	1019	0.87 ± 0.1	0.90 ± 0.05

During this period of time, several sodium samples were obtained for the purpose of determining chemical and radioactive components. Samples were obtained with a dip tube technique using three stainless steel cups, each with a capacity of approximately 16 g. Following the acquisition of the sodium sample, and allowing a sufficient time for the decay of the sodium-24 component, the sample was dissolved in methyl alcohol, and a 2-ml aliquot was transferred to a liquid scintillation vial for tritium determination. A comparison of these results with the calculated tritium concentration from the ternary fission yield is shown in Table 1.

It is noted that the tritium production in the sodium results primarily from the ternary fission process, and that it is in agreement with calculations. From these data, it would appear that a large fraction of the tritium produced as a result of ternary fission is released into the sodium coolant.

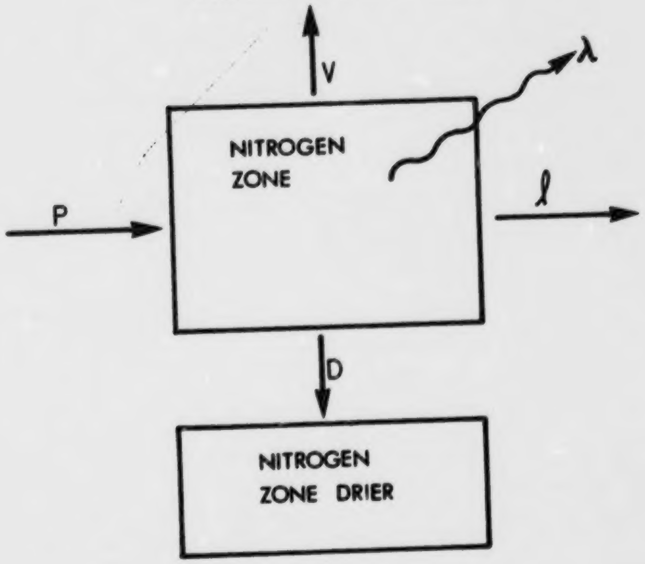
No data are presently available upon the effect of cold trapping the tritium at SEFOR, as was noted of the EBR-II in a recent paper by Ebersole et al. [5]. It has not yet been possible to examine in detail the distribution of tritium throughout the SEFOR sodium systems. These measurements are planned when the resumption of power and the completion of other test activities will permit.

Comparison of Calculated Data in the Nitrogen Zone

A simple model, Figure 2, was employed to described the concentration of tritium in the nitrogen zone in which the venting, leakage, tritium decay and removal by condensation are balanced by the production at equilibrium, i.e.:

Figure 2

Tritium Concentration Model



$$P_T = \lambda N + lN + DN + VN$$

$$\lambda N = \frac{\lambda P_T}{\lambda + V + l + D} = \text{equilibrium activity in the nitrogen zone}$$

where

N = total number of tritium atoms in nitrogen zone

P_T = production rate of tritium (4.0×10^{10} atoms/sec)

V = fraction of tritium atoms vented from N_2 zone/sec (1.2×10^{-7} sec^{-1})

l = fraction of tritium atoms leaking from N_2 zone/sec (5.2×10^{-8} sec^{-1})

λ = decay constant of tritium (1.79×10^{-9} sec^{-1})

D = fraction of tritium atoms circulated to drier (7.0×10^{-5} sec^{-1})

Then $\lambda N = 1.03 \times 10^6$ dps for 20 MW operation of 28 μCi at equilibrium. The concentration in the nitrogen zone then becomes 28 μCi/16.8 X 10^8 ml or 17 pCi/l at equilibrium 20 MW operation. Since the rate of produc-

Table 2

Calculated Tritium Levels in Nitrogen Drier Condensate

Date	EFPH	Increment EFPH	μCi	nCi/ml
12/11	460.2	-	-	-
12/18	500.0	40.2	280.0	9.3
12/19	501.0	1.0	7.0	0.23
12/28	560.0	59.0	420.0	1.4
1/4	600.0	40.0	280.0	9.3
1/8	670.0	70.0	490.0	16.0
2/17	940.0	270.0	1890.0	62.0

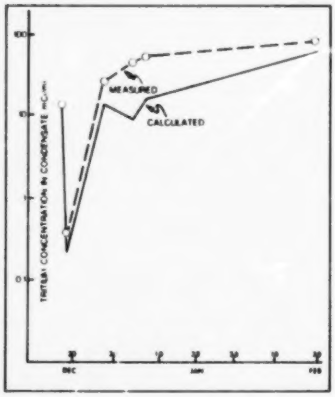
Table 3

Measured Tritium Levels in Nitrogen Drier Condensate

Date	Corrected nCi/ml
12/18/70	13.8
12/19/70	0.36
12/28/70	26.4
1/4/71	42.5
1/8/71	54.0
2/17/71	8.3
2/20/71	2.4

Figure 3

Tritium Concentration in Nitrogen Zone Drier Condensate



tion is balanced by the rate of removal, the input to the nitrogen zone drier is 4.0 X 10^{10} atoms/sec.

For one EFPH, 1.44 X 10^{14} atoms of tritium are transferred to the drier, corresponding to 7.0 μCi. Equilibrium conditions in the nitrogen zone are achieved after approximately 18.2 hours. Assuming a 100 percent efficiency for the removal of the tritium atoms from the nitrogen zone, the input to the drier is thus 7.0 μCi for each EFPH.

For a period of approximately two months, samples of the nitrogen zone drier condensate were removed at frequent intervals. These were analyzed for tritium content, and the results are shown in Tables 2 and 3. A comparison of the estimated and the measured concentration of tritium in the condensate is presented as Figure 3.

Application and Health Physics Significance

Although the production rate of tritium in the nitrogen zone as a result of fast neutron interaction with nitrogen and the boron component of the serpentine rods results only in a few millicuries a year, tritium has constituted the most significant source of liquid radioactive waste at the SEFOR Facility. This is not due to the concentration of tritium in the nitrogen zone itself, which is of slight health physics significance, but is primarily a result of the highly efficient collection and retention of tritium in the condensate of the nitrogen zone drier. The water collected in the drier is periodically pumped to the liquid radioactive waste system where the concentration of tritium frequently requires dilution to conform to waste disposal criteria. It is interesting to note that this source of tritium may have some health physics significance in a large Liquid Metal Fast Breeder Reactor, typically of a power perhaps 100 times the power of the SEFOR reactor.

While minimization of tritium production in Liquid Metal Fast Breeder Reactors has not been a design objective, proposed reactor designs which utilize an enclosure of the entire primary system in a large vessel remove the need for an extensive nitrogen atmosphere. Hence, this source of tritium production becomes of less importance. For a pressure vessel design, the source of tritium from fast neutron irradiation of nitrogen will be dependent upon neutron leakage from the pressure vessel.

ACKNOWLEDGMENTS

The review and suggestions offered by J. O. Arterburn, Manager, SEFOR Facility, are gratefully appreciated. Appreciation is also expressed to Dr. Mitty C. Plummer for his help and suggestions.

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AIRBORNE TRITIUM MONITORING NEAR A HEAVY WATER NUCLEAR REACTOR

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A portion of the Argonne Environmental Monitoring Program deals with tritiated water vapor in air. The measured tritium distributions on the Argonne National Laboratory (ANL) site are consistent with the interpretation that the CP-5 reactor is the only significant localized tritium source in the area. The ANL CP-5 reactor is a heavy-water moderated and cooled research reactor with a maximum power output of five megawatts. The core contains about 10^4 liters of 99.4% D_2O, which has a tritium concentration of 1.52 mCi/ml. In the normal mode of operation, the concentration of tritium in the exhaust air is about 2 μCi/m^3. Essentially all of this tritium is in the form of tritiated water vapor.

Collection and Measurement

The tritiated water vapor was collected by adsorption onto silica gel (6 to 16 mesh) contained in a basket. The basket is about 10 X 10 X 2 cm, constructed of 14 mesh copper screening and includes a hook for supporting the basket in the field. One hundred grams of silica gel were weighed out for each basket, dried at 400°C for two hours and then stored at 110°C until needed.

Baskets with silica gel were placed in the field for periods ranging from one to three days. As part of the routine monitoring program, two sample collection baskets were maintained near the CP-5 reactor at all times. One basket was placed 50 meters east of the reactor, while the second basket was placed about 100 to 200 meters from the reactor in the most favorable location for the detection of tritium based on the prevailing wind speed and direction. The baskets were changed every other day during most of the study. In addition to these baskets, other samplers were placed at various locations on and off the ANL site, but not on a continuous basis, to map tritium distributions.

Several studies were made with an environmental test chamber to measure the rate at which water is adsorbed by silica gel. The chamber provided both constant temperature and humidity for extended periods of time. A set of silica gel samplers was placed in the test chamber at a known temperature and humidity and then removed sequentially at selected times. The volume of water collected by each sampler was measured and plotted as a function of time. Under the various combinations of temperature and humidity tested, the curve was essentially linear for the first 20 hours with an adsorption rate of 1 ml/h. For times greater than 20 hours, the adsorption rate decreased, and the curve asymptotically approached the saturation level of water on silica gel. Although these data indicate that samples should be changed daily for the best results, this could not be done because of manpower limitations.

Immediately after the sample basket was brought in from the field, the silica gel was transferred to a Pyrex tube and placed in a furnace at 400°C until all the water was removed. This required at least 30 minutes. The water that was driven off was condensed and collected. The elbow between the tube and condenser contained a coarse porosity sintered glass disc, which served as a trapping device for impurities and eliminated a quenching problem in counting tritium that had previously existed.

The tritium concentration was determined by liquid scintillation counting using a modification of the Triton N-101 gel system developed by Moghissi et al. [1]. The scintillation solution contained 7.0 g 2,5-diphenyloxazole (PPO) and 1.35 g p-bis(o-Methylstyryl)-Benzene (bis-MSB) dissolved in and diluted to one liter with toluene. This solution was mixed with 500 ml of N-101 to produce the scintillation mixture. A 15-ml aliquot of this solution was dispensed into 10 ml of sample in a polyethylene vial, shaken well and allowed to gel overnight before counting was started.

Counting was done in a Packard Instrument Co. Model 3320 liquid scintillation spectrometer. The refrigerator which contained the samples was maintained at 6°C. Previous work showed that at 12°C, toluene loss from the samples was significant after a few days and at 4°C, the gel took several days to set. At 6°C, the samples would gel overnight and remain stable for several weeks.

Samples were counted for 100-minute periods, with a minimum of three counts per sample. The tritium counting efficiency was about 20%, and the blank values were 13 counts per minute. The efficiency was measured by adding a known amount of tritium to a second aliquot of about 10% of the samples. Under these conditions, the limit of detection is about one cpm, or 0.5 dpm/ml. The counting results in units of tritium activity per milliliter of water were converted to activity per cubic meter of air with the aid of the temperature-humidity data from a recording hydrothermograph operated continuously near the CP-5 reactor.

Discussion

Any discussion of the concentration of tritium in the environment due to a localized source must be prefaced by a discussion of the ambient levels from widespread sources of the existing tritium. This level of tritium is made up of the contribution from natural sources (cosmic-ray induced nuclear reactions, fission, neutron reactions, etc.) and nuclear tests.

To ascertain the ambient or base line tritium concentration in air, a number of far off-site samples were taken at various locations. These averaged about 2 pCi/m^3 during the last six months of this study. The specific activity of tritium in these samples was approximately 500 pCi/l. Tritium concentrations in surface water and precipitation in the Chicago area were also measured regularly during this study as part of the Environmental Monitoring Program at ANL. The average tritium concentration in surface water was also about 500 pCi/l. The tritium content in monthly composite precipitation samples is given in Table 1. With the exception of the increase in April and May, 1971, which was probably due to a spring increase in stratospheric fallout, and the exceptionally high value in October, 1970 probably due to an atmospheric nuclear test, these concentrations were also similar to those of surface water. Thus, surface water, precipitation, and water vapor have approximately the same tritium content, giving credence to the conclusion that the ambient tritium concentration in air is about 2 pCi/m^3. Fluctuations in air concentrations about this value are expected due to changing meteorological conditions. Periodic off-site sampling has been continued to monitor this base line value.

Table 1

Tritium in Rain

Month	Av. pCi/l
July, 1970	464
August, 1970	742
September, 1970	288
October, 1970	2335
November, 1970	531
December, 1970	711
January, 1971	288
February, 1971	655
March, 1971	678
April, 1971	999
May, 1971	1064
June, 1971	594

Although sampling for tritium in air has been a continuing program for the last several years, the discussion will concern itself primarily with data collected since July 1, 1970. A summary of this data for the area near the CP-5 reactor is presented in Table 2. Considerable fluctuation exists between individual samples,

Table 2

Tritium Concentrations Near the CP-5 Reactor

Month	No. of Samples	Tritium Concentration (pCi/m^3) Av.	Min.	Max.
July, 1970	43	452	18	2680
August, 1970	32	556	10	2510
September, 1970	26	319	4.5	1590
October, 1970	26	160	2.2	1240
November, 1970	24	201	5.4	980
December, 1970	24	94	1.2	433
January, 1971	26	213	1.4	1126
February, 1971	24	267	1.8	1160
March, 1971	26	355	5.7	1565
April, 1971	26	331	5.9	1631
May, 1971	26	234	5.2	2148
June, 1971	12	847	20.8	4084
Summary	315	336	1.2	4084

due largely to variations in wind direction and humidity/temperature changes during the sampling period. In addition, rain is very effective at lowering the tritium concentration in air containing tritiated water at above ambient concentrations.

Samples were also collected twice a month in the East Area, 2000 meters northeast of the reactor. These data are presented in Table 3. In these samples the tritium concentrations ranged from 0.55 to 32 pCi/m^3 and averaged 8.5 pCi/m^3. The East Area samples generally varied in the same manner as those taken near the reactor when the prevailing wind direction and velocity are considered. In general, East Area samples were greater than 10 pCi/m^3 on days when samples at the CP-5 reactor were greater than 100 pCi/m^3 and the wind was from the southwest. The correlation is sufficiently good to conclude that most of the tritium in the East Area came from the reactor.

Samples were collected twice a month from a control point located about ten kilometers northwest of the ANL site. The results are given in Table 3. The tritium concentrations ranged from 0.23 to 13 pCi/m^3 and averaged 3.5 pCi/m^3. No correlation between high results at the CP-5 reactor and off-site results could be determined.

Table 3

Tritium Concentrations Away from CP-5

	March 10, 1971	March 17, 1971	March 24, 1971
Wind Direction*	180°-45° (S-NE)	330°-180° (NW-S)	315° (NW)
CP-5	328	578	891
East Area	5.3	3.3	2.2
A	4.3	2.5	2.0
B	5.4	1.6	1.6
C	1.8	1.3	2.4
D	3.9	2.1	1.3
E	-	1.6	1.9

East Area - 2 kilometers NE of CP-5.
A - 4 kilometers E of CP-5.
B - 5 kilometers SW of CP-5.
C - 10 kilometers NW of CP-5.
D - 16 kilometers N of CP-5.
E - 24 kilometers E of CP-5.

*Direction from which wind was blowing.

Table 4

Tritium Concentrations in Off-Site Air Samples (pCi/m^3)

	East Area[1]				Off-Site[2]			
Month	No. of Samples	Conc. (pCi/m^3) Av.	Min.	Max.	No. of Samples	Conc. (pCi/m^3) Av.	Min.	Max.
July, 1970	1	-	-	31	1	-	-	6.3
August, 1970	2	24	16	32	2	12	10	13
September, 1970	2	16	14	18	2	3.8	3.0	4.6
October, 1970	2	4.2	3.3	5.1	2	6.4	3.7	9.0
November, 1970	2	21	4.6	37	2	3.6	2.5	4.8
December, 1970	2	4.7	1.2	8.1	2	2.1	1.4	2.8
January, 1971	2	1.4	0.55	2.3	2	0.30	0.23	0.37
February, 1971	2	5.1	4.3	5.8	3	2.2	1.0	4.1
March, 1971	4	3.0	1.3	5.3	17	2.3	1.0	5.4
April, 1971	1	-	-	2.2	1	-	-	2.0
May, 1971	2	13	4.2	22	2	5.4	3.6	7.2
June, 1971	1	-	-	7.2	1	-	-	9.2
Summary	23	8.5	0.55	32	37	3.5	0.23	13

[1] Two kilometers northeast of the reactor.
[2] Ten kilometers northwest of the reactor.

In an effort to determine if any tritium could be detected off the ANL site in any direction, samples were collected at various distances on three occasions; results are presented in Table 4. The choice of sampling stations was limited to secure locations. Inspection of the results shows that all off-site samples had tritium concentrations similar to the base line data, and there was no positive correlation between concentration and wind direction. There is no indication that any tritium produced by the CP-5 reactor was found off the ANL site.

In another set of experiments, baskets were placed on the site approximately 600 meters from the CP-5 reactor at the four compass points. These data, presented in Table 5, show a very strong correlation between sample location and wind direction at 600 meters. Application of these results to a mathematical model met with limited success. In a two-dimensional system, the concentration should fall off inversely as the square of the distance from the source $(1/d^2)$. In a linear system, where the tritium is carried in one direction by the wind, the decrease in concentration should be proportional to the reciprocal of the distance (1/d). The best fit to the experimental data was obtained when the exponent of d was 1.25. This would imply the tritium is carried by the wind with some, but limited, conical distribution. A rigorous analysis is not possible because of the limited data and the complexity of airborne distributions.

To assess tritium concentrations in terms of radiological safety, they may be compared to the environmental maximum permissible concentrations (MPC) specified by the International Commission on Radiological Protection for members of the public. This maximum permissible concentration is 200 nCi/m^3, so the average concentration near the CP-5 reactor was 0.17% of the MPC. For samples collected at greater distances, the fraction of MPC is correspondingly lower. Thus, the ambient tritium concentration of about 2 pCi/m^3 is 0.001% of the MPC.

Table 5

Tritium Concentrations in On-Site Air Samples
(picocuries per cubic meter)

	May 7, 1971	May 14, 1971	May 21, 1971	July 13, 1971
Wind Direction[a]	45°-120° (NE-SE)	60°-135° (NE-SE)	190° (S)	270°-300° (W-NW)
CP-5	398	240	89	325
600 Meters North	14	20	178	23
600 Meters West	44	87	6.9	13
600 Meters South	22	14	4.5	146
600 Meters East	7.8	14	5.6	56

[a] Direction from which wind was blowing.

Conclusion

In this study, a method is presented to continuously monitor the airborne release of tritiated water vapor. The sample collection, preparation, measurement, and calculation techniques are discussed, and results of the program are presented. The experimental data show that the concentration of tritium in air falls off quite rapidly with distance from the source; it was not detectable above background tritium levels off the ANL site.

ACKNOWLEDGMENTS

This work was performed under the auspices of the U. S. Atomic Energy Commission.

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TRITIUM GENERATION AND RELEASE TO IN-PLANT AND OFF-SITE ENVIRONS OF THE LA CROSSE BOILING WATER REACTOR

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The La Crosse Boiling Water Reactor (LACBWR) is a 165 MW(th) forced circulation, direct cycle boiling water reactor, located on the east bank of the Mississippi River approximately 19 miles south of La Crosse, Wisconsin. Constructed by Allis-Chalmers Manufacturing Company, the reactor is owned by the Atomic Energy Commission and is operated under prime contract by Dairyland Power Cooperative. Dairyland owns the 50 MW electric turbine generator which is connected to the Dairyland Power grid.

The reactor operates at a pressure of 1300 psia. Water returning from the condenser and feedwater heaters at 285°F is mixed with recirculating coolant and heated to 577°F to supply approximately 600,000 lb/h of steam to the turbine generator. Initially, the reactor core consisted of approximately 8600 kg of 3.63 percent ^{235}U enriched UO_2 pellets clad in 0.020-inch 348-H stainless steel. There are 72 fuel assemblies, each having 100 fuel rods. Reactor control is achieved through the use of 29 cruciform control rods which contain B_4C. The reactor vessel is constructed of 4-inch thick type A302B carbon steel clad with 0.19 inches of type 304 L stainless steel. The surface condenser tubes are 0.040-inch thick admiralty. Both stainless steel and carbon steel are utilized in the primary system piping.

The LACBWR achieved criticality in July 1967, was first connected to the DPC grid in April 1968 and reached full design power in August 1969. After repair of eleven sensitized stainless steel safe ends in the primary system in late 1969 and early 1970, and successful operation in 1970, the plant was declared commercial in February 1971. As of July 1971, fuel burnup has reached approximately 6000 MWD/MT with no apparent cladding defects. Stack release rates for activation and fission gases have averaged less than 100 μCi/sec. Measurements of tritium in LACBWR systems began in 1970.

A multitude of information concerning tritium has been summarized by Jacobs [1]. Additional information concerning tritium as it affects nuclear power is documented in recent hearings before the Joint Committee on Atomic Energy, Ninety-first Congress [2]. Information on tritium has also been reported by Kahn et al., in a study at Dresden I [3] and in a survey paper by Peterson [4]. Tritium production rates from fission have been well documented and are summarized by Horrocks [5]. This paper will discuss mechanisms of release of the tritium produced at the La Crosse Boiling Water Reactor as measured during the last two years of operation, in addition to other aspects of tritium at the facility.

Measurement of Tritium at LACBWR

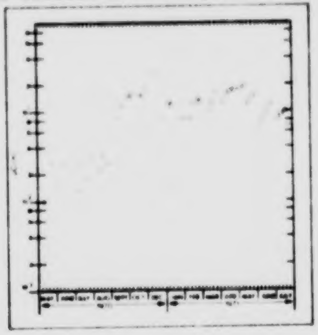
Tritium is measured in water as tritiated water, utilizing a nonrefrigerated single sample liquid scintillation counter. Calibrations have been performed using tritium standards and intercomparisons with other laboratories. Concentrations of tritium in air are determined by cold trapping moisture from the air and counting it utilizing a dioxane, napthalene, PPO, and POPOP cocktail. Air concentrations can be determined through dew point measurement. Tritriated water concentrations have also been measured by commercial laboratories, whose sensitivities are 1 pCi/ml.

Tritium in the LACBWR Primary Systems

The tritium concentration in the LACBWR primary system (reactor vessel) has been measured weekly, Figure 1. The tritium concentration

Figure 1

Concentration of Tritium in Primary Water as a Function of Time



is a function of power level and system integrity. The concentrations in the main steam and feed water have been analyzed and, within the statistical accuracy of the analysis, these are equivalent.

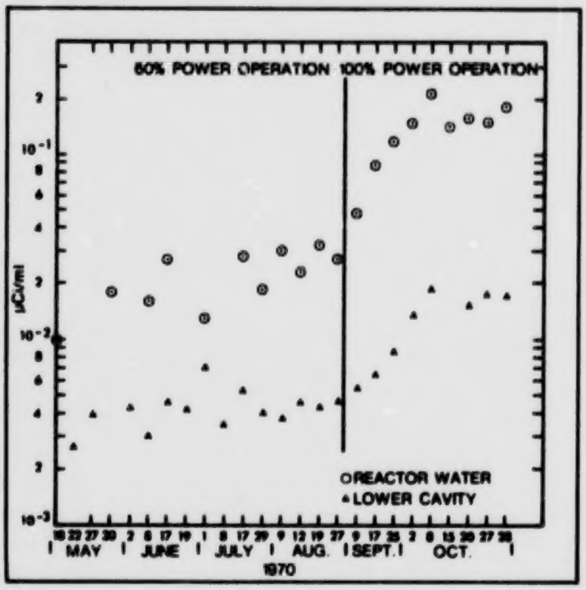
Use of Tritium as a Leak Indicator

Since tritium is in sufficient concentration in the LACBWR primary system, it is used as a sensor of primary system leakage. Using an activity balance, leaks as small as 1 cc/min can be detected in the LACBWR reactor vessel and associated piping. A previous LACBWR report [6] discussed the relationship between the concentration of tritium in the reactor water and the condensed water vapor in the closed air space surrounding the reactor vessel and piping. The air space contains only piping and vessel, with no valves. These data are plotted in Figure 2. A linear relationship was not indicated by the data. A diffusion model is more nearly represented where the

$$\frac{\left(\begin{array}{c}\text{concentration of tritium in}\\ \text{reactor vessel at 100\% power}\end{array}\right)^{1/2}}{\left(\begin{array}{c}\text{concentration of tritium in}\\ \text{reactor vessel at 60\% power}\end{array}\right)^{1/2}} = \left(\frac{\begin{array}{c}\text{concentration of tritium in}\\ \text{reactor cavity at 100\% power}\end{array}}{\begin{array}{c}\text{concentration of tritium in}\\ \text{reactor cavity at 60\% power}\end{array}}\right)$$

Figure 2

Concentration of Tritium in Primary Water and Lower Cavity Condensate as a Function of Time



Since the presence of tritium outside of the reactor vessel and piping was indicated by a diffusion phenomena, other pathways to the environment were suspected.

Monitoring for tritium in various areas of the plant gives a good indication of primary system leakage from valves, etc. This is especially true where routine entry cannot be made due to the radiation exposure received during inspection of high radiation areas. In addition, when defective fuel is not present, tritium is frequently the only indicator of leakage.

Tritium Balance at LACBWR

Table 1 indicates the amount of tritium produced, calculated on the basis of 0.30 tritium atoms/10^4 fissions. Tritium production from control rods and deuterium activation was not considered, because of the small contribution they make in comparison to ternary fission.

Each month the amount of tritium released in liquid wastes is determined by analyzing each waste batch. Stack tritium releases were determined quantitatively during 1971 by cold trapping water vapor for tritium from the stack gas. The amount released for the month has been

Table 1

LACBWR Tritium Produced and Released

Month	MWd(th)	Tritium* Produced (Ci)	Tritium Released (Ci)			Remarks
			Liquid Waste	Stack	Condenser Cooling Water	
May '70	1586	16.17	0.43			Limited to 60% Power
Jun '70	1468	14.97	0.95			Limited to 60% Power
Jul '70	2610	26.62	0.54			Limited to 60% Power
Aug '70	2910	29.68	0.16			Limited to 60% Power
Sep '70	3885	39.63	6.04			Escalated to 100% Power
Oct '70	4539	46.30	4.37			100% Power Operation
Nov '70	S/D	-	2.57			Shutdown for Maintenance
Dec '70	1661	16.94	3.64			85 to 100% Power Operation
Jan '71	3253	33.18	4.5	6.7		85 to 100% Power Operation
Feb '71	3325	33.92	4.5	2.4		85 to 100% Power Operation
Mar '71	3181	32.45	6.5	2.4		85 to 100% Power Operation
Apr '71	3740	38.15	5.1	3.6	11	85 to 100% Power Operation
May '71	3885	39.63	8.6	3.3	25	85 to 100% Power Operation
Jun '71	3938	40.17	14.5	2.2	None Detectable	85 to 100% Power Operation

*From ternary fission only

calculated from the average of the weekly grab samples. During April, May and June 1971, samples of the surface condenser inlet and outlet were sampled to determine possible tritium diffusion through the admiralty condenser tubes. The amount released has been calculated from weekly grab samples.

Discussion of Tritium Release Pathways

The amount of tritium released in liquid waste is mostly a function of primary water tritium concentration. Comparing operation at 60 percent power to operation at 100 percent power, it can be noted that releases are a function of primary water concentration rather than the integrated power or the amount of tritium produced from fission. Tritium liquid waste releases per month have varied from 0.5 percent of the amount produced at 60 percent power to 36 percent of the amount produced at 100 percent power.

As can be noted from the stack release of tritum during January 1971, a leak above normal did exist. Examination of the data from tritium air samples taken in the plant indicated the leak was from the turbine generator portion of the facility. This leak, which was in a turbine inlet valve, was repaired during February 1971, and the amount released showed a decrease.

Table 2

Tritium Release From the Stack

SAMPLE NO.	3	4	5	6	7	8	9	10	11	12	13	14	15
DATE	4/7/71	4/14/71	4/21/71	4/28/71	5/5/71	5/12/71	5/19/71	5/26/71	6/2/71	6/11/71	6/17/71	6/23/71	6/30/71
COND. FLOW (GPM)	30,000	30,000	30,000	30,000	30,000	30,000	30,000	30,000	30,000	60,000	60,000	60,000	60,000
TEMP. IN °F	40	48	51	49	51	56	61	52	61	68	77	75	78
TEMP. OUT °F	59	70	71	70	73	80	82	76	80	80	88	87	90
PRIMARY ³H µCi/ml	0.1207	0.155	0.167	0.173	0.177	0.191	0.122	0.163	0.155	0.0725	0.0893	0.0980	0.118
COND. VACUUM IN Hg	28.55	28.20	27.95	27.50	27.67	27.65	27.4	28.1	27.70	28.00	27.66	27.67	27.56
CONDENSER INLET TRITIUM CONCENTRATION X 10^{-6} µCi/ml	0.0±1.0	0.0±0.01	0.0±1	0.0±1	0.0±1	0.0±1	0.0±1	0.0±1	0.0±1	0.0±1	0.0±1	0.1±1	0.0±1
CONDENSER OUTLET TRITIUM CONCENTRATION X 10^{-6} µCi/ml	1.7±0.4	0.1±0.1	4.0±0.6	1.1±0.1	0.5±0.9	0.0±1	1.5±0.6	0.0±1	12.0±1.1	0.0±1	0.0±1	0.0±1	1.9±0.4

Samples taken from the surface condenser cooling water inlet and outlet in April and May 1971 indicated a release of tritium across the condenser on ten of twelve samples. The detailed conditions at the time of sampling are noted in Table 2. During this period, the inlet temperature varied between 40° and 61°F and the outlet temperature between 59° and 82°F, with half the maximum condenser cooling water flow (30,000 gallons per minute). The condenser vacuum during this period ranged between 27.4 and 28.6 inches of mercury, and the primary tritium concentration ranged from 0.122 to 0.177 µCi/ml. Only one of four samples taken in June 1971 indicated an increase between the influent and effluent of the condenser. During this period of time, the condenser inlet temperature ranged from 68° to 78°F and the outlet from 80° to 90°F. Also, the condenser cooling water flow had been increased from 30,000 to 60,000 gallons per minute.

The mechanism of release from the surface condenser cannot be conclusively determined from the data in Table 2. However, it is apparent that from 30 to 60 percent of the amount produced can be released to the environment through this pathway. Additional investigation into this release mechanism is necessary to quantify the amount released.

In-Plant Tritium Radiation Exposures

The concentration of tritium in air at LACBWR is measured routinely by cold trapping water vapor for analysis. Concentrations in air have varied from $<10^{-9}$ μCi/ml to 2.5×10^{-7} μCi/ml. They are a function of system leak tightness and primary tritium concentration. The International Commission on Radiological Protection (ICRP) and Federal Radiation Council and hence, AEC, have recommended that the maximum permissible tritium air concentration for a 40-hour week be 5×10^{-6} μCi/ml. The leakage from LACBWR has never indicated that the concentration is greater than 5 percent of this guide.

Urine samples have been obtained from LACBWR employees routinely entering these areas (occupancy factor <5 percent) and have indicated tritium in urine in the range of 20 to 40 pCi/ml. The resultant calculated internal dose to employees from tritium is less than 5 mrad/year. This is insignificant when compared to the average external radiation dose received by the personnel and with the accepted ICRP and NCRP recommendations.

Estimated Doses to Off-Site Environs from Tritium Releases

The primary mode of exposure to the off-site environs from a direct cycle boiling water reactor is from the cloud gamma dose from the plume released from the stack. As can be noted from the data, tritium releases from LACBWR are predominantly liquid releases. Tritium releases from the stack are less than 0.1 percent of permissible levels based on LACBWR meteorology and stack height. Releases in the liquid waste and to the condenser are also less than 0.1 percent of recommended ICRP guides.

Neither of these pathways to the environment results in a radiation exposure to surrounding populations greater than 1 mrad/year. This dose cannot be measured, and it would only be academic to calculate the exact dose. Hence, tritium is not of immediate concern, from a dose standpoint, to populations surrounding the LACBWR.

Conclusions

A large percentage of tritium produced in a direct cycle boiling water reactor utilizing stainless steel cladding, such as the LACBWR, is released to the primary water and plant environs and eventually to the environment. The percentage of the tritium produced by ternary fission which is released at LACBWR is mostly a function of the primary water concentration and, hence, the amount diffusing through the fuel cladding. The primary pathway to the environment is liquid waste and possible condenser cooling water. During recent months of LACBWR operation, from 50 to 95 percent of the tritium produced from ternary fission was released if equilibrium conditions are assumed. The diffusion mechanism appears to be the primary pathway of leakage to the environment. Additional investigation is necessary to both quantify and determine the release mechanism of tritium through the surface condenser.

Internal radiation doses from tritium to operating personnel are a small percentage of the total dose received in the operation and maintenance of the LACBWR. Off-site radiation doses cannot be measured. Calculations indicate they are an extremely small percentage of natural background radiation and hence do not constitute any hazard to the health and safety of the public.

Tritium can be used effectively as an indicator of primary system integrity and can assist the operator of a nuclear power facility in determining the location and amount of a leakage before it becomes an operational problem.

ACKNOWLEDGMENTS

The author wishes to acknowledge the assistance of R. Craig, J. Shannon, D. Larson and L. Krajewski in the collection and analysis of the samples referred to in this report.

This report was prepared as an account of work sponsored by the United States Government. Neither the United States nor the United States Atomic Energy Commission, nor any of their employees, nor any of their contractors, subcontractors, nor their employees, make any warranty, express or implied, or assume any legal liability or responsibility for the accuracy, completeness or usefulness of any information, apparatus, produce or process disclosed, or represent that its use would not infringe privately-owned rights.

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ENVIRONMENTAL TRITIUM SURVEILLANCE FOR PROJECT RULISON

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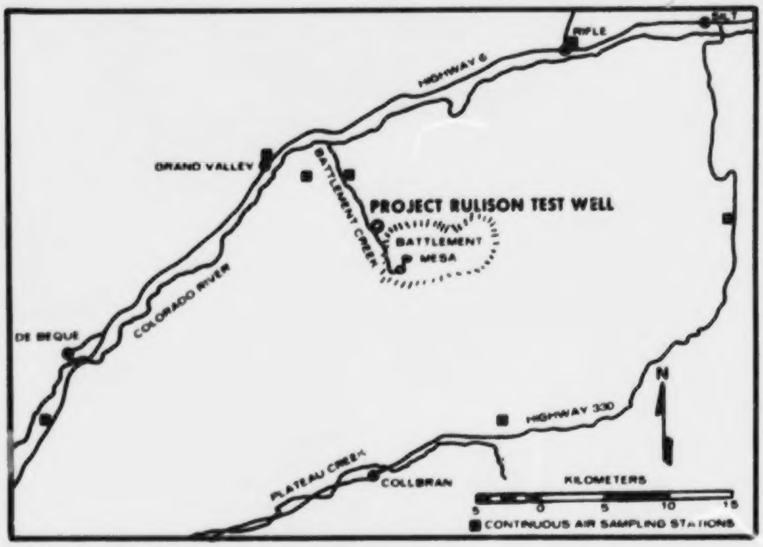
Project Rulison was the second nuclear experiment conducted jointly by the U.S. Government and private industry to investigate the feasibility of using nuclear explosives to stimulate production of natural gas from a low-permeability gas-bearing formation by producing a zone of fractured rock and a chimney of rock rubble around and above the detonation point. The void spaces thus created provided a reservoir into which the natural gas could flow. A standard gas well was then drilled to the chimney, allowing recovery of the gas. This technique was shown to be successful in the first such experiment, Project Gasbuggy, conducted by the U.S. Government and El Paso Natural Gas Company near Farmington, NM, in December 1967 [1]. Project Rulison was conducted by the U.S. Atomic Energy Commission, Department of Interior, and Austral Oil Company near Grand Valley, CO. The nuclear device was detonated at a depth of 2570 m below the ground surface in September 1969, and chimney reentry and production testing of the test well extended from April 1970 through April 1971.

In accordance with a Memorandum of Understanding between the Environmental Protection Agency and U.S. Atomic Energy Commission, the Western Environmental Research Laboratory (formerly the Southwestern Radiological Health Laboratory of the Public Health Service) conducted an off-site radiological surveillance program for the detonation, chimney reentry, and production testing phases of the project. The detonation phase was completed with no release of radioactivity to the environment [2,3]. Reentry operations to drill a well into the area of rubble and fractured rock near the detonation point began in April 1970. By the time the area of broken rock was reached in July, most of the gaseous radioactivity originally produced by the detonation had decayed to stable nuclides. Essentially all of the nongaseous radioactivity was expected to have been trapped in the solidified rock which melted during detonation, or in the chimney debris. Consequently, tritium and ^{85}Kr were the two remaining radionuclides of primary interest expected to be released to the environment as part of the natural gas that was to be burned, or flared, at the surface during production flow tests on the test well. It was assumed that after burning, any tritium present in the plume would be in the form of water vapor.

The off-site surveillance program for the reentry and production test phases was, therefore, designed to monitor the environment for tritium and ^{85}Kr in particular. The program was also designed to monitor for ^{14}C and other radioactive products that might be released from the test well. Because of the relative ease of collecting atmospheric moisture for tritium analysis and the somewhat greater public health significance expected for the released tritium, it was chosen as the radionuclide for routine air surveillance. Net ^{85}Kr concentrations could be estimated from net tritium concentrations and from the measured ratio of the two gases at the wellhead. In addition, tritium would be the only radionuclide expected in other environmental samples. This paper reports the procedures and results of the off-site tritium monitoring program conducted during the reentry and production testing phases by the Western Environmental Research Laboratory (WERL).

Figure 1

Project Rulison Area



Background

The Project Rulison test well is located in a mountain valley approximately 64 km northeast of Grand Junction, CO. The nearest community (Grand Valley, population 300) is 10 km northwest of the site along the Colorado River. Figure 1 shows a map of the area. A 27.4-m flare stack was erected about 100 m from the test well to burn the natural gas released during reentry and production testing operations. The wellhead is at an elevation of 2,480 m above mean sea level in the upper end of Battlement Creek valley. Battlement Creek is one of several streams draining the slopes of Battlement Mesa, a 3,290-m flat-topped mountain.

Battlement Creek has no flowing tributaries and originates from several ponds and reservoirs on top of Battlement Mesa, 3 to 5 km southeast of the project site. The stream then flows within 100 m of the wellhead, and continues northwest where it empties into the Colorado River at an elevation of 1,570 m near the town of Grand Valley. About 5 km downstream from the wellhead, the creek leaves the valley and crosses an alluvial fan area known as Morrisania Mesa.

Several ranches are scattered across Morrisania Mesa, many of which use Battlement Creek for irrigation and domestic water. The Clem Ranch, located near the mouth of Battlement Creek valley, is the nearest ranch to the test well and served as one of the primary surveillance stations in the area. Battlement Creek valley and Battlement Mesa are uninhabited except for occasional hunters, fishermen, and ranchers tending stock. The Colorado River and its tributaries drain the north side of Battlement Mesa. Plateau Creek and its tributaries drain the southern side, eventually emptying into the Colorado River about 35 km downstream from Grand Valley. Various ranches and the communities of Grand Valley, Rifle, Silt, Collbran, Plateau City, and Mesa are located along these two waterways.

At the higher altitudes, the predominant wind direction across the Battlement Mesa area is toward the northeast. Occasional winds blow to the east or southeast, but rarely in other directions [4]. Wind directions near the surface are influenced by the terrain and other factors, such as temperature inversions and nocturnal cooling. The predominant wind conditions in the vicinity of the test well were of particular importance in terms of where the plume from the gas flaring stack would be carried. The predominant daytime condition involved up-slope winds in the Battlement Creek valley, allowing the plume to rise and be carried over the 2,700- to 3,000-m ridges immediately east and southeast of the test well. From there, the plume remained primarily under the influence of winds at those levels, although vertical mixing to lower altitudes occurred under some conditions. Plume travel to the nearest populated location under these conditions was usually 16 km or more. Nocturnal flow under cloudy sky conditions usually remained up-slope under the influence of prevailing winds. With clear sky conditions, a nocturnal down slope or drainage wind in the Battlement Creek valley was common. The nighttime down-slope drainage wind was the normal occurrence. Under these conditions, a portion of the plume would be carried down the valley to the Morrisania Mesa area, while another portion would rise through the drainage wind layer to higher altitudes and be carried to the east.

Drilling operations began on April 28, 1970, to reenter the chimney and establish a well so gas production tests could be conducted. The initial phase of this operation involved redrilling a well about 100 m from the nuclear device emplacement well, angled to intercept the chimney slightly above the detonation point. Special drilling equipment and techniques were used to assure no gas was released from the well during drilling operations.

Once the area of broken rock was reached, the well was connected to water and drilling mud separator systems on the surface. The material thus collected from the gas was stored in tanks to await disposal. The gas stream was checked for radioactivity and other constituents by several sampling and monitoring systems before routing to the flare stack, where it was burned in the atmosphere. As water accumulated in the tanks of the separator circuits, it was periodically injected into the gas stream to be vaporized in the flare.

The test well was completed on July 28, 1970, and the first gas release occurred on August 1. Several short calibration flaring runs were scheduled thereafter to determine plume trajectories, dispersion parameters, and off-site radioactivity concentrations for several gas flow rates before full-scale production testing began. Operational problems were encountered when the gas lines became plugged with mud and rock debris from the well. Although the calibration tests were consequently cut short by this and other difficulties, sufficient information was gathered to indicate that production testing could proceed. Following remedial measures to reopen the well, production test flaring began in the fall of 1970. The various flaring operations of production testing through April 1971 are summarized in Table 1.

Table 1

Summary of Project Rulison Flaring Operations

Flaring Period	Date	Flow Rate $10^6 Ft^3/d$	Date	Flow Rate $10^6 Ft^3/d$	Volume Gas Flared $10^6 Ft^3$
Preliminary Flow Tests*	8/1/70	0.8-17.0	8/22/70	-	1
Calibration Tests**	10/4/70	2.0-15.0	10/7/70	-	12
High-Rate	10/27/70	17.0	11/3/70	11.4	109
Intermediate Rate	12/1/70	5	12/20/70	5	100
Long-Term	2/2/71	11.5	4/23/71	0.9	233

*Nine short runs up to 8 hours' duration.

**Three short runs up to 14 hours' duration.

Operational Procedures

The off-site environmental tritium monitoring program consisted of three basic efforts. These were (1) aerial plume tracking and sampling using a WERL fixed-wing aircraft, (2) special intensive atmospheric moisture sampling surveys during calibration flaring and selected periods of the long-term flaring, and (3) regular sampling of air, water, milk, natural vegetation, soil, crops, animal tissue, and urine throughout the reentry and production testing period. All samples were returned to the WERL in Las Vegas for analysis, with the exception of some stream and atmospheric moisture samples analyzed at a temporary field laboratory in Grand Junction, CO, during the initial calibration flaring runs. Routine samples were shipped daily to Las Vegas by commercial air transportation. Aerial and other special samples were returned immediately to Las Vegas after collection on WERL aircraft. Analytical data were transmitted to Colorado for field operation guidance and to Project

Rulison Open Files maintained in Las Vegas, Denver, and Bartlesville, OK, by the Atomic Energy Commission for data dissemination to the public.

Aerial plume tracking and sampling were performed with a twin-engined Turbo-Beech aircraft. Since radioactivity levels in the plume from the gas flaring stack were so low that gamma and beta detectors could not be used, plume tracking was accomplished with a condensation nuclei monitoring instrument, manufactured by Environment/One Corporation of Schenectady, NY. Output from this typewriter-sized instrument, which measured the airborne concentration of condensation nuclei resulting from burning of the natural gas, was fed to a strip-chart recorder in the crew chief's instrument panel, providing him with continuous information on plume trajectory and size. This information was radioed to a ground control center so ground monitoring personnel could be positioned to collect samples in the plume ground track.

Atmospheric moisture samples were collected through the aircraft sampling probes with a cryogenic sampling system and a grab sampler. The cryogenic sampler consists of a series of traps submerged in a sealed 22-l dewar filled with liquid nitrogen. A pressure relief valve maintains a pressure of approximately 0.77 kg/cm^2 on the liquid nitrogen, resulting in a temperature of about $84^{o}K$. Water and CO_2 are frozen out in the initial traps or removed as snow in two consecutive cyclone separators. The noble gases are collected in a final trap containing 13X molecular sieve. A $3-m^3$ sample was collected over a 30-minute period while the aircraft circled or spiraled in the plume.

Grab samples were obtained by gathering air from the sampling probe in a $1-m^3$ plastic bag over a 30-second period, and pumping the air at 60 l/min through a cannister containing 1000 g of Linde type 13X molecular sieve for water vapor and CO_2 collection; it was then stored in a compressed air bottle.

Prior to designing the various atmospheric moisture samplers used in surveillance for Project Rulison, a study of collection characteristics was conducted at the WERL. Collection efficiencies of 97 percent or greater were observed with absolute humidities of 15 to 30 ml of water per cubic meter of air as long as flow rates remained below 60 l/min per 300 g of molecular sieve. Bed capacity was about 20 percent by weight before breakthrough occurred. The molecular sieve samplers were designed so that bed residence times were at least 25 percent greater than the minimum given above. Depending on temperature and humidity, the molecular sieve samplers were about 25 to 50 percent efficient for CO_2. Only a portion of the CO_2 collected was required for ^{14}C analysis. The ^{14}C concentration in the air was calculated from the concentration in CO_2 and the reported CO_2 atmospheric concentration of 0.03 percent. The grab sample volume of one cubic meter was chosen to allow carrier-free separation of Kr and to provide sufficient water for tritium analysis.

The bottled air and cryogenic sampler were returned to the WERL for ^{85}Kr analysis. Water and CO_2 recovered from the molecular sieve and cryogenic sampler were used to determine tritium and ^{14}C concentrations.

Based on aerial tracking information, monitoring personnel were instructed to drive to ground stations along roads or were airlifted by helicopter to otherwise inaccessible areas in the downwind trajectories. The monitors were located at predesignated sampling stations and carried portable two-way radios to receive instructions concerning sampling times and position changes.

Portable battery-powered air samplers were used to collect atmospheric moisture samples. A Bon Aire automobile vacuum cleaner (Lear Siegler, Inc.) served as the air mover with power supplied by a 12-V automobile battery. Air was drawn through a 10.2-cm diameter glass fiber particulate filter followed by a 300-g cartridge of molecular sieve at a flow rate of about 45 l/min. Sample volume was measured by a Rockwell dry gas meter. All dry gas meters had been calibrated against the WERL 14-m^3 spirometer and were assigned individual calibration factors. Sampling time was controlled to collect 10 to 20 ml of water, based on field calculations of absolute humidity. Humidity measurements were made with sling psychrometers. Airborne tritium concentrations were based on the sample volume and total tritium in the water vapor collected. Portable battery-powered compressors and a truck-mounted cryogenic sampler were also used to collect air samples for tritium and ^{85}Kr analysis. Carbon-14 analysis was performed on the CO_2 from representative molecular sieve and cryogenic samplers.

Continuous collection of atmospheric moisture during flaring periods was accomplished at seven fixed monitoring stations at populated locations in the vicinity of the test well, Figure 1. Metal storage sheds were set up to house the samplers. Air was drawn through a particulate filter outside the sampling shed, then through a cannister containing 700 g of molecular sieve for water collection. The sampler operated for 48-hour periods at a flow rate of 3 l/min, controlled by a limiting orifice. The total volume of air sampled each period was calculated from the flow rate and operating time. Absolute humidity was determined from the total volume of water collected.

During the initial flaring operations, dehumidifier units developed by the Eastern Environmental Radiation Laboratory of the EPA were also operated at each station to collect atmospheric moisture samples over 24-hour sampling periods. Standard room dehumidifiers were modified with the addition of a heating strip inserted in the cooling coils and a manually adjustable timer. The dehumidifier samplers operated in the cool cycle for 25 minutes, during which atmospheric moisture was frozen out on the cooling coils. A five minute heat cycle melted the accumulated ice, which flowed into a plastic collector. Temperature and humidity data at each station were recorded continuously by hygrothermographs housed in standard meteorological instrument shelters located at least 3 m from the sampling sheds. Absolute humidity calculated from recording hygrothermograph data was used with counting data from the dehumidifier samples to determine atmospheric tritium concentrations if any positive samples were obtained. These samples were counted for tritium with a special scintillation flow cell unit at the Grand Junction field laboratory. The dehumidifier samplers provided a means of collecting atmospheric moisture samples at any given time for a rapid assessment of atmospheric tritium concentrations. Daily water samples from Battlement Creek were also counted in the scintillation flow cell counter during initial flaring operations.

Precipitation collectors were provided at each of the seven fixed sampling stations, and portable precipitation collectors were deployed by field personnel at other locations as conditions dictated. For precipitation collection, a funnel with an open area of 530 cm^2 was placed on the roof of each sampling shed.

Precipitation was routed from the funnel to a graduated cylinder inside the shed. Portable precipitation samplers consisted of plastic sheets held 0.3 m above the ground on a 1-m by 1-m frame. The water collected was drained into a container for shipment to the laboratory. Several snow samples were collected with this device, and many other grab snow samples were collected during the winter flaring operations.

Continuous water samples were collected from two stations on Battlement Creek with battery-powered automatic water samplers (Brailsford and Co.). One sampler was located about 100 m down stream from the test well, and the second about 5 km downstream, near the mouth of Battlement Creek valley and just upstream from the first withdrawal point for domestic water supplies. Daily 3.8-liter samples were collected in this manner until October 7, 1970, when the samplers had to be removed due to freezing weather. After that, a daily grab sample was collected at the lower station during flaring operations. All other water sampling stations were sampled on a monthly schedule. These included 10 streams, four reservoirs, six wells, four springs, and 15 municipal supplies.

Milk samples from 11 family cows and four Grade A dairies were also collected on a monthly basis for tritium analysis. Garden foods were sampled when available. Natural vegetation and domestic animal forage were also sampled monthly, when available. Periodic samples of animal tissue from both wildlife and domestic animals were collected through the assistance of local meat packing houses and the Colorado Fish, Game and Parks Department. Several blood samples were collected from domestic livestock for tritium analysis when tissue was not available from a particular area of interest or at a particular time of interest (for example, livestock near the mouth of Battlement Creek valley immediately following a flaring operation). Urine samples were collected from 20 residents in the area during flaring operations, when the maximum tritium concentration could be expected if exposure occurred. All samples were returned to the WERL for analysis.

Analytical Procedures

Water recovery procedures at the laboratory for the various types of samples were as listed in Table 2. All samples analyzed for tritium in Las Vegas were counted in a Beckman LS-100 liquid scintillation counter [5]. With a minimum sample volume of 5 ml, the minimum detectable tritium concentration is 0.4 pCi/ml of water.

The scintillation flow cell counting system used at Grand Junction was developed at the WERL on the basis of work reported by Burt and Gibson [6]. A spiral flow cell filled with anthracene crystals was optically coupled to a photomultiplier with output to a scaler-timer. Voids volume in the cell was 0.8 ml. Due to the high background, approximately 0.5 mR/h, the minimum detectable concentration of tritium was 600 pCi/ml of water. Although this was far above the normally expected concentrations for atmospheric moisture or water from Battlement Creek, it provided a wide margin of safety as a rapid analysis tool for public protection. With normal absolute humidities of less than 5 ml of water per m^3 of air, a detection limit of less than

Table 2

Analytical Techniques

Type Sample	Processed by
Molecular Sieve	Heat and vacuum degassing
Water	Thermal distillation
Milk	Thermal distillation
Urine	Thermal distillation
Vegetation	Freeze-drying
Animal Tissue	Freeze-drying
Soil	Freeze-drying

3 nCi/m^3 was provided, compared to the allowable exposure to a suitable sample of the population, averaged over a year, of 67 nCi/m^3. A satisfactory margin of safety was provided, also, for surface water with an allowable concentration of 1 nCi/ml averaged over a year.

Results of tritium analysis performed in Grand Junction were available the day the sample was collected. All data and work sheets were transmitted by Telecopier to Las Vegas on a daily basis for inclusion into the Project Rulison Open Files.

Results

Samples of atmospheric moisture, water, milk, precipitation, natural vegetation, soil, food crops, bovine feed, animal tissue, and urine were collected prior to any release of gas and analyzed to determine the background levels of tritium in each type of sample. The range of background sample results and the highest above-background results associated with flaring operations are shown in Table 3.

All samples collected by the WERL were collected off-site (greater than 180 m from the flare stack) except for tissue from two porcupines which grazed directly under the flare stack for several days. Tissue from these porcupines contained elevated levels of tritium as high as 180 pCi/ml of water. The levels in the tissue water were still below the Radioactivity Concentration Guide (RCG) for tritium in water for the general off-site population (1 nCi/ml of water) [7]. All other animal tissue and blood samples collected during the flaring operations contained background levels of tritium.

All samples of milk, water, food crops, and bovine feed collected during and after the flaring operations also contained background levels of tritium. Some samples of atmospheric moisture, precipitation, natural vegetation, and soil collected in the off-site area contained concentrations of tritium greater than background. These elevated concentrations were observed in generally isolated instances during flaring operations, and were not observed as continuing elevated levels of any duration, either

Table 3

Environmental Levels for Tritium in the Project Rulison Area

Sample Type	Background Range (pCi/ml H_2O)	Highest Above-Background Result (pCi/ml H_2O)
Atmospheric Moisture	0.5 - 2.6	59 ± 1.0
Water (stream)	0.4 - 1.6	None
Water (well)	0.4 - 1.0	None
Water (spring)	0.4 - 1.4	None
Water (all)	0.4 - 1.6	None
Milk	0.4 - 2.1	None
Precipitation	0.4 - 2.1	47 ± 0.9
Natural Vegetation	0.4 - 5.8	21 ± 0.6
Soil (surface)	0.4 - 1.4	6.7 ± 0.4
Soil (15 cm depth)	0.4 - 1.1	None
Soil (all)	0.4 - 1.4	6.7 ± 0.4
Food Crops	0.4 - 3.1	None
Bovine Feed	0.7 - 4.0	None
Animal Tissue	0.6 - 1.5	180 ± 1.7
Urine	0.4 - 1.1	None[*]

[*] Urine from several individuals in the area indicated tritium concentrations ranging to 9.5 pCi/ml, but these above-background values were shown to result from wearing tritium-activated luminous dial watches, not Project Rulison activities [8].

during or after a flaring operation. Urine samples from several residents in the area indicated tritium concentrations ranging to 9.5 pCi/ml, but these above-background values were shown to result from wearing tritium-activated luminous dial watches, not Project Rulison activities [8].

The highest tritium concentration in an atmospheric moisture sample collected during the flaring was 59 ± 1 pCi/ml of water, equivalent to 290 ± 5 pCi/m^3 of air. This was less than one percent of the RCG for the general off-site population (67 nCi/m^3 of air). This sample was collected 1.3 km downwind from the flare stack in an unpopulated location on Battlement Mesa during a calibration flaring period. Slightly elevated levels of tritium were also observed in populated areas around the flare stack. The highest concentration in an atmospheric moisture sample collected at a populated location was 11 ± 0.5 pCi/ml of water, or 21 ± 0.9 pCi/m^3 of air. This is less than 0.1 percent of the off-site RCG. This sample was collected at the closest populated location (about 5 km) to the flare stack. Elevated levels of tritium were also detected in DeBeque, Grand Valley, Rifle, and Collbran for short periods of time, see Figure 1.

The highest tritium concentration in a precipitation sample collected during the flaring operation was 47 ± 0.9 pCi/ml of water. It was a snow sample collected about 200 m from the flare stack during a wintertime flaring operation. This is less than five percent of the off-site RCG for tritium in water. The highest concentration of tritium in a precipitation sample collected from a populated location was 2.5 ± 1.1 pCi/ml of water in a rain sample collected 7 km northwest of the flare stack. This concentration of tritium is not significantly greater than the maximum background concentration of 2.1 ± 1.1 pCi/ml and is less than one percent of the off-site RCG.

A vegetation sample collected from the top of Battlement Mesa immediately following a calibration flaring operation contained the highest tritium concentration detected in vegetation. This was 21 ± 0.6 pCi/ml of water, or 7.6 ± 0.2 nCi/kg wet weight. The highest tritium concentration detected in a vegetation sample collected at a populated location was 3.0 ± 0.3 pCi/ml of water, or 1.3 ± 0.1 nCi/kg wet weight. This sample was collected near Silt, CO.

A surface soil sample collected 0.4 km northwest of the test well contained the highest tritium level detected in soil of 6.7 ± 0.4 pCi/ml of water. The highest tritium concentration detected in a soil sample collected at a populated location was 2.8 ± 0.3 pCi/ml of water at Collbran.

Discussion

Elevated tritium concentrations in atmospheric moisture and ^{85}Kr were found to be the most likely, and possibly the only, sources of population exposure from Project activities. Slightly elevated concentrations of tritium found in natural vegetation, soil, and precipitation were from a few isolated samples from scattered locations, and represented no widespread contamination that could provide a significant source of exposure to man. No concentrations above background were observed in water, milk, food crops, or animal tissue (domestic livestock or wildlife) that could be consumed by man. The only elevated tritium concentrations found in animal tissue occurred in two small herbivores that grazed extensively on a small patch of vegetation that remained green within a few meters of the flare stack during an extended wintertime flaring operation. This patch remained green as a result of heat generated by the flare, while the surrounding area was covered by snow and all other vegetation was dormant. The tritium in the forage eaten by these animals was probably primarily due to uptake from the rain-out of moisture which condensed from the plume in the immediate stack vicinity. Other wildlife samples (such as deer and elk) collected within several kilometers of the flaring stack gave no indication of elevated tritium levels.

Since this surveillance showed that atmospheric moisture was the only source of tritium exposure to man, the monitoring data from these samples were used to estimate a possible dose from the flaring operations. Estimates were prepared for each of the continuous monitoring stations operated at populated locations, including the one nearest the test well at the mouth of Battlement Creek valley. The net average concentrations for each sampling period were integrated over the respective sampling periods, summed for all periods, and then converted to dose using standard conversion parameters [9]. These estimates showed the maximum dose to a resident in the Project Rulison area from exposure to tritium was less than 0.001 mrem. Urine samples collected from residents showed no measurable exposure from Project Rulison activities.

Although no increase in urine tritium concentrations were detected as a result of Project Rulison flaring operations, concentrations ranging up to ten times background were measured before and during flaring in persons wearing tritiated wrist watches. The estimated average annual dose to wearers of the watches ranged from 0.2 to 1.8 mrem. The WERL is continuing its investigation of this finding and details of the wrist watch study are to be reported soon [7].

NOTICE

Mention of brand names does not imply endorsement by the Environmental Protection Agency.

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EXPERIMENTAL RESULTS FROM PROCESSING GASBUGGY GAS IN A NATURAL GAS PROCESSING PLANT

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The use of nuclear devices to increase natural gas production from otherwise unproductive gas fields has been tested in the Gasbuggy and Rulison experiments. Other tests are planned. The use of such nuclear devices results in low levels of tritium and ^{85}Kr in the natural gas produced from such wells.

Studies in the Health Physics Division of the Oak Ridge National Laboratory (ORNL) are providing estimates of the potential radiation dose that might occur through the use of gas produced from nuclearly stimulated wells [1-3]. The experiment described in this paper was performed in cooperation with El Paso Natural Gas Company (EPNG) to test the validity of prior calculations [1] on the distribution of tritium among the various processing plant products ("dry" gas, butane, propane, and natural gasoline), to determine if isotopic exchange occurs between tritiated components of natural gas and the absorption oil used for product extraction, and to study the exposure of plant operating personnel during the processing of gas from a nuclearly stimulated well. Results of the experiment are given in greater detail in an ORNL report [4].

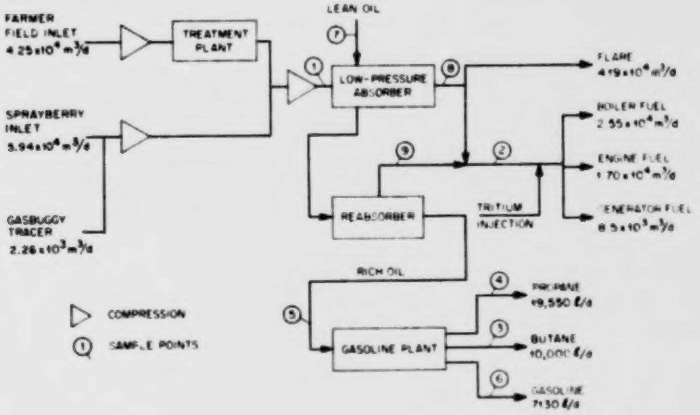
Natural gas from the Gasbuggy well, developed by a 29-kiloton nuclear device detonated 1280 meters underground on December 10, 1967, near Farmington, NM, was transported to the test site in Texas in two propane tank trucks containing 1130 cubic meters of gas. The Gasbuggy Project was a joint undertaking by the U.S. Atomic Energy Commission, the U.S. Bureau of Mines and El Paso Natural Gas Company to evaluate the feasibility of the use of nuclear explosives to stimulate low productivity oil and gas reservoirs.

This 1130 cubic meters of tritium contaminated gas was blended with 5.1×10^4 cubic meters of field gas and processed in a natural gasoline plant during a 12-hour period as shown in Figure 1. During the Gasbuggy gas flow period and the subsequent 12 hours, the plant liquid products were diverted to storage (and later burned on-site) and the processed gas was used to fuel compressors, generators, and boilers in the plant, or was flared. No tritium contaminated natural gas went to customers, although the mixed inlet gas stream contained only 0.22 to 0.27 pCi/ml of tritium.

Because of the low tritium concentration, it was necessary to supply additional tritium to the plant fuel gas as shown in Figure 1. To achieve this, approximately 70 mCi of tritium in elemental form (furnished by the ORNL Isotopes Division) was transferred from its shipping container into a small gas cylinder with 700-psi plant gas

Figure 1

Simplified Diagram of Barnhart Plant Showing Test Flow Rates and Sample Points



and metered into the fuel gas stream during the test.

Sampling and Analysis

Samples of the plant gas streams were obtained in one-liter steel bombs. The high-vapor-pressure, condensed liquid streams were sampled in smaller bombs. Natural gasoline and lean absorption oil samples were taken in glass bottles. Thirty weighed desiccant samplers were distributed at head height throughout the plant to absorb atmospheric moisture during the test period. These consisted of approximately 200 g of indicating Drierite contained in mesh screens. The samplers were kept in sealed containers before and after use. Temperature and humidity readings were made at intervals during the test so that atmospheric tritium concentrations could be calculated from the tritium content of the moisture samples.

The area moisture samples were vacuum distilled from the desiccant at ORNL, and 1 ml of water from each sampler was counted for several 50-minute periods in a liquid scintillator. The samples were then spiked to measure scintillator efficiency. Large variations in efficiency were noted, so the remaining portion of each sample was distilled from Na_2O_2. This resulted in normal scintillator efficiency. The sample for atmospheric tritium background correction was taken in the same manner as the other samples in an area free from gas combustion products.

The natural gasoline samples were counted as sampled, with the initial sample taken as zero. All other liquid samples were burned over copper oxide and counted as liquid water. The instrumental background sample was doubly electrolyzed water considered to be tritium free.

This sample gave 1.05 cpm. All samples received at least ten 50-minute counts. The gas samples were counted in a proportional counter with selected samples analytically separated into the gaseous components which were counted individually.

All samples were chemically analyzed to determine the mole percent of each constituent. We believe that the tritium concentration in those samples burned to produce liquid water is reliable to at least ±1 pCi/ml and the concentration in the gross gas samples is reliable to at least ±10%. Difficulties that we experienced in achieving reproducible results lead us to believe that routine application of analytical procedures to low-level tritium samples without careful quality control is likely to result in large uncertainties.

Results

The total inventory of tritium in the transported Gasbuggy gas was calculated to be 12.38±1 mCi, distributed as shown in Table 1.

Table 1

Composition of Gasbuggy Tanker Gas and Tritium Distribution

Component	Composition (mole %)	Tritium Content (mCi)
CH_4+H_2	75.0	8.81
C_2H_6	8.2	2.34
C_3H_8	8.5	0.49
C_4H_{10}	3.0	0.49
$C_5H_{12}^+$	0.9	0.25
Inert	4.4	

After mixing with field gas, the average resulting concentrations during the 12-hour test are shown in Table 2.

Table 2

Composition of Mixed Plant Gas

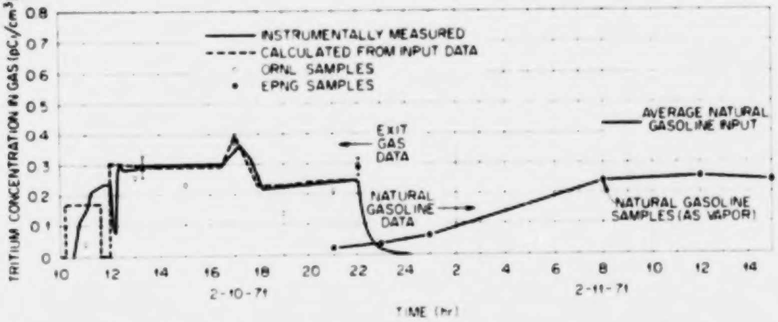
Component	Composition (mole %)	Tritium Concentration pCi/ml	
		Pure Component	Total Gas
CH_4+H_2	77.4	0.224	0.173
C_2H_6	8.6	0.535	0.046
C_3H_8	6.2	0.261	0.010
C_4H_{10}'s	2.5	0.400	0.010
$C_5H_{12}^+$	1.2	0.417	0.005
Inert	4.1		
TOTAL			0.245

The values for propane appear anomalous. The high percentage of this component and its low tritium concentration resulted from the fact that the transport tankers contained one atmosphere of uncontaminated propane, their normal cargo, when the Gasbuggy gas was loaded.

The values shown in Table 1 were calculated from three analyses of tanker gas taken during the test, plus fractionation data from higher hydrocarbons (liquids) separated from the gas at the Gasbuggy well. The values in Table 2 result from hydrocarbon composition data on incoming field gas during the test, combined with Table 1 data weighted by a material balance on plant products. To compare calculated and observed data, refer to Figure 2. Here actual recorded data from the El Paso Natural Gas calibrated, on-line monitor is compared to the calculated value for tritium concentration in the exit gas. It can be seen that the on-line monitor follows variations in dilution faithfully with the expected time lag and lack of definition caused by in-plant time constants.

Figure 2

Tritium Concentration in the Most Volatile and Least Volatile Plant Product Streams vs. Time



On the right side of Figure 2, the gradual appearance of the tritium associated with the natural gasoline product stream is shown. These concentrations are expressed as pCi/ml of gasoline vapor for comparison purposes. This product is liquid at ambient conditions, consisting mainly of a mixture of pentanes, hexanes, and heptanes. In a plant operating at design capacity, this product would show tritium contamination much more rapidly than we observed, since this plant was operated at less than 15% of design capacity during the experiment.

The fact that the actual tritium concentration did not attain the calculated pure component value stems from plant operational limits. The liquified products (propane, butane, and natural gasoline) are absorbed from the incoming gas stream into absorption oil (No. 1 diesel oil) and then distilled as a mixed liquified fraction into a receiving tank that feeds further columns which separate the mixed product into propane, butane, and natural gasoline. This series of mixing processes results in effective dilution of tritium in the product streams since process equilibrium was not attained by the end of the 12-hour Gasbuggy gas flow period.

During the test, samples were taken of all product streams and at several sampling points within the processing system. It is beyond the scope of this paper to review these data in detail. In general, the observed distribution of tritium within the processing system and among the plant products confirmed theoretical calculations of the expected distribution of tritium among the various separated hydrocarbons, providing confidence in our ability to predict quantities of radioactivity in gas and liquid products leaving a plant that processes gas from nuclearly stimulated wells.

All of the liquid products are absorbed and desorbed from absorption oil. Additional oil is supplied at a rate of 0.5 to 1% per day to replace processing losses. If isotopic exchange of tritium in the processed natural gas and hydrogen in the absorber oil occurs, a considerable inventory of tritium would build up in the plant, although at concentrations not substantially greater than those observed in the natural gasoline fraction. Prior experiments at ORNL with tritiated methane and ethane saturating kerosene at high pressure for 30 to 60 days showed no exchange under conditions similar to those existing during the plant test [5].

To confirm these observations under actual process conditions, samples were taken during the test of both rich (gas saturated) and lean (product stripped) absorption oil. The results are shown in Table 3. The rich and lean oil results were determined on the water formed by oxidation over copper oxide. Although the values are small, it can be seen that the lean oil remains essentially tritium-free. If as much as 1% of the tritium inventory had exchanged with the estimated 6 X 10^4 liters of absorption oil, the lean oil would have contained ≈2 pCi/ml of activity. It was concluded that no gross exchange occurred during the test and perhaps none, confirming the results of the above-mentioned ORNL experiments.

Table 3

Tritium Content of Rich and Lean
Absorption Oil Samples

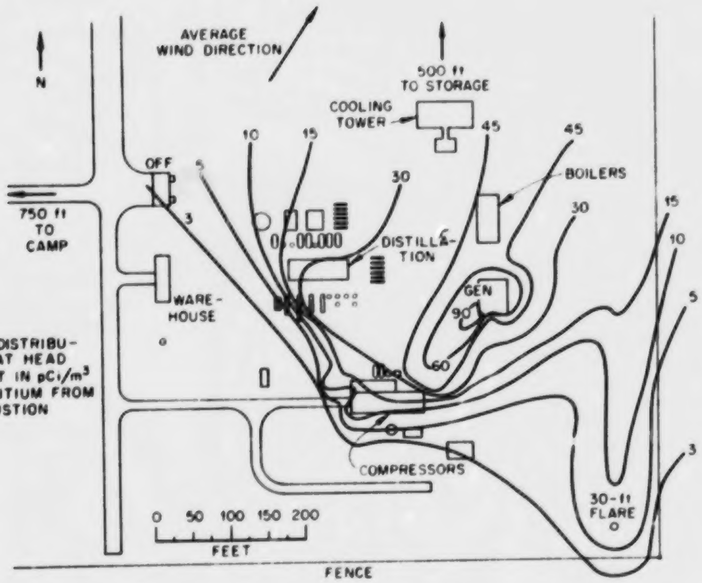
Sample Time	Tritium Concentration (pCi/ml)		Vapor from rich oil
Date Hour	Lean Oil	Rich Oil	(Gross β)
2-10-71, 1235	--	<1.4	0.14
2-10-71, 1300	-0.40	1.44	--
2-10-71, 1700	+0.73	--	--
2-10-71, 1855	--	8.2	0.33
2-10-71, 2100	--	2.27	--
2-10-71, 2227	--	2.5	0.30
2-11-71, 0800	-0.48	4.6	--
2-11-71, 1200	-0.63	--	--
2-11-71, 1300	-1.13	--	--

Exposure of Plant Operating Personnel

To assess the potential exposure of plant workers to the combustion products from fuel-burning plant equipment (compressors, generators, and boilers) and to be able to compare field results

Figure 3

Area Distribution (at Head Height) of Tritium from Gas Combustion Products (Picocuries of Tritium per Cubic Meter of Air)



with calculated values from previous studies, approximately 70 mCi of tritium in elemental form was added to the plant fuel gas.

The highest measured tritium content of the fuel gas stream was 6.1±0.2 pCi/ml during the test and the average concentration was determined to be ~4.0 pCi/ml. The tritium concentration in the water collected by our samplers ranged from 0.4 to 32.3 nCi/ml of water. After applying atmospheric temperature and humidity data to calculate average pCi/m^3 of tritium as tritiated water over the test period, it was possible to plot isobars of concentration on a plot plan of the gasoline plant as shown in Figure 3.

During the test period, a gusty wind from the southwest blew at about 15 to 30 miles per hour. This resulted in the combustion effluent from the short compressor stacks on the south side of the compressor building downwashing into the area around the generator building. The plant product gas feeding the flare contained Gasbuggy tracer at less than 0.3 pCi/ml of ^{3}H, so no large effect was noted downwind of the flare. The plant auxiliaries were burning gas at a rate of 1.8 X 10^6 ft^3/day, which is consistent with the usage in a much larger gasoline plant operating on product from a high-pressure natural gas field requiring less fuel for compressors. It is unlikely that more than ten times this fuel usage would occur in any plant.

Table 4

Tritium Dose Commitments Resulting From Exposure During Plant Test

Organization	Individual	Date Samples	Total Dose Commitment (millirem ±0.02)
EPNG	A	2/11	0.65
	B	2/11	0.13
	C	2/10	0.10
	C	2/11	0.14
ORNL	A	2/10	0.35
	A	2/11	0.36
	B	2/11	0.05
	C	2/10	BG
USBM	A	2/11	BG
Eberline	A	2/11	0.17
	B	2/11	0.08
Truck	A	2/10	BG
	B	2/10	BG
EPNG	1	2/10	BG
	1	2/11	0.026
	2	2/10	BG
	2	2/11	0.029
	3	2/10	0.024
	3	2/11	BG
	4	2/10	BG
	4	2/10	BG
	5	2/10	BG
	5	2/11	BG
	6	2/11	BG
	7	2/10	0.035
	8	2/11	BG
	9	2/10	BG
	10	2/10	BG
	11	2/10	BG
	12	2/10	BG
	13	2/11	BG
	14	2/10	BG
	15	2/10	BG
	16	2/10	BG

A conservative estimate of maximum potential annual dose commitment may be made by multiplying the highest concentration (131 pCi/m^3) within the 90 pCi/m^3 isobar in Figure 3 by ten. Using 0.126 mrem/day as the dose commitment from 1 μCi body burden of tritium and 2000 hours of exposure/a, a dose commitment of 0.6 mrem/a is found, which would not appear an unwarranted risk for an occupational worker. In comparison, it is estimated that the El Paso Natural Gas Company Blanco Plant, which processes 576 million cubic feet of gas per day and burns 11×10^6 ft^3/day on-site, would have an average ground-level concentration of 50 pCi/m^3 when processing 1.1 pCi/ml gas, or ≈300 pCi/m^3 on an equivalent basis to the conservative estimate.

In addition to area moisture samples, urine samples were taken from personnel involved in the test program. These samples were returned to ORNL where the tritium concentration in each sample was determined by liquid scintillation counting. After counting for 20 minutes, the average background count of these samples was 13 cpm, and the random scatter may be as much as 1.3 cpm/ml. With a counting efficiency of >0.15 (measured), a precision of about ±8.8 cpm/ml is realized. This corresponds to ±4 pCi/ml of urine.

Essentially no correction was necessary for decay time since ingestion with these samples. The measured results were converted to body burden using the parameters of standard man [6]. The resulting dose commitment was then calculated assuming 1 μCi of tritium body burden delivers 0.126 mrem.

The uncertainty in the calculated dose is ±0.02 mrem plus the deviations of the individual from standard man. The results are given in Table 4 with calculated doses less than 0.02 mrem reported as background. In the table, technical and supervisory personnel are identified by letters and operators by numerals.

The four people involved in the transfer of the 70 mCi of tritium into a gas cylinder (including three authors) received dose commitments ranging from 0.13 to 0.65 mrem and values from 0.08 to 0.17 mrem were found for the on-line instrument calibrators.

Doses calculated for inhalation and skin absorption using the air concentrations determined from the area moisture samples are much lower than even the small operational doses shown. The only credible mechanism for these doses is direct leakage from equipment exhausts within the plant buildings.

ACKNOWLEDGMENTS

Thanks are due to H. M. Butler and O. M. Sealand, ORNL, who assisted in the performance of the experiment; D. G. Jacobs, ORNL, who helped in the pretest planning; Jerry Abrams, U.S. Bureau of Mines, Bartlesville, who operated the USBM on-line tritium monitor; A. E. Doles, R. L. Powell, and L. M. Phyfe, Eberline Instrument Corp., who operated the EPNG on-line monitor; Don Johnson, plant superintendent, and Jim Cronenberg, EPNG, who assisted in the preparations for the test and in its performance; and Carl Mullins, Texas State Health Department, who made observations concerning the radiological safety of the test.

This research was sponsored by the U.S. Atomic Energy Commission under contract with the Union Carbide Corporation.

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HEALTH PHYSICS ASPECTS OF A LARGE ACCIDENTAL TRITIUM RELEASE

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On Thursday morning, August 6, 1970, 289 kCi of elemental tritium were accidentally released through the stack (36.5 meters high) of the Gaseous Chemistry Building at the Lawrence Livermore Laboratory (LLL) in Livermore, CA. Shortly after the radiation alarm was sounded, emergency response personnel were called out to operate tritium air monitors and to collect environmental samples in order to assess the immediate impact of the release.

During the following four days, an extensive environmental sampling program was undertaken by the Hazards Control Department of the LLL with the assistance of personnel from the California State Health Department. Numerous air, water, vegetation, milk, and urine samples were collected and analyzed for tritium. The survey covered an area extending northeast from Livermore Valley to Lodi, CA, and southeast in the San Joaquin Valley from Lodi to Visalia, CA. This sampling pattern was based on the meteorological conditions that prevailed during and shortly after the release. The emergency response activities immediately following the release are described in this report and the results obtained from the environmental sampling program are compared with pre-accident measurements and off-site maximum permissible concentrations (MPC) as defined in the U.S. Atomic Energy Commission Manual (Chapter 0524, "Standards for Radiation Protection").

The Sequence of Events

The release of the tritium was detected and the amount recorded by a flow-through ionization chamber that sent a high-level radiation alarm to the LLL Fire Station at 0614 on Thursday, August 6, 1970. A spill team was activated at 0615 and the Off-Site Emergency Survey Team was called in at 0708 to operate tritium air monitors and to collect environmental samples on-site and along several roads north and east of the Laboratory. Predictions based on early meteorological data indicated that the activity would be carried to the northeast by light southwesterly winds.

At 0802 a urine-sampling program was initiated for all personnel involved in the initial emergency response. Urine samples were also obtained from other LLL employees who were on-site during the release. An extensive air, water, vegetation, and on-site urine sampling program was continued throughout the day.

In the initial off-site survey, environmental samples were collected along the roads between Livermore and Tracy, and southeast of Tracy through the San Joaquin Valley as far as Firebaugh. Since the release involved the dispersal of radioactivity to the off-site environment, a representative of the California State Health Department was notified at 1530, Thursday, August 6. This representative then assisted in the collection of milk samples, which were returned to LLL for analysis on Friday, August 7.

Results of the first day's environmental sampling program were summarized at 0800 the following morning (August 7). Vegetation samples containing low levels of tritium activity had been collected at a few locations northeast of the Laboratory; based on these results, environmental sampling was increased in this area. Additional samples were collected around Tracy and farther north at Lodi.

On Friday afternoon (August 7), an off-site urine sampling program was initiated after consultation with the representative of the California State Department of Public Health. Based on meteorological predictions and initial vegetation sampling results, it was found that the area immediately north and northeast of LLL was exposed to the highest tritium concentration during cloud passage. Urine samples were therefore obtained from several individuals who were in this area at the time of release. The off-site environmental sampling program was continued for several weeks, with primary emphasis on those areas where tritium was detected in vegetation samples.

Meteorological Predictions

The atmospheric conditions at LLL at the time of release were characterized by south-to-southwest winds at 1 to 2 m/sec. Morning radiosonde data from the Oakland Airport indicated a surface-based inversion with a top at about 850 m. This inversion probably prevailed over the Livermore Valley. Tritium was released into the atmosphere at an effective height of about 50 m, and surface winds would have carried the cloud northward, see Figure 1, where it would have been forced into various canyons in the area about one hour after release, i.e., at 0730. At this time, the cloud may have been forced upward into the predominantly westerly flow that existed at the higher altitudes according to wind data from the Laboratory's Site 300. In any case, the cloud was forced over the foothills in the Altamont Pass Road area and into the San Joaquin Valley. Best estimates of arrival time at Byron Road would be 0900 to 1000. At this time, surface winds at Tracy changed from light southeast to strong westerly, which indicated the possible breakup of the inversion. The cloud would probably have moved slightly northward during its eastward passage, passing north of Tracy at approximately 1100.

Stockton and Manteca surface winds were northerly to northwesterly at 3 to 6 m/sec between 1100 and 1300. As the cloud moved closer to Stockton, it would have been forced southward and probably passed between Tracy and Manteca at about 1200, continuing on a southerly track down the San Joaquin Valley. Using the surface winds from Modesto, Merced, and Crows Landing, the cloud could be estimated to have crossed Patterson Road (J-17) at about 1400 and reached the Los Baños area at about 1600.

Environmental and Bioassay Measurements

The environmental survey program consisted of direct air monitoring and the collection of water vapor, vegetation, water and milk samples. Each of these survey techniques is discussed in the following paragraphs.

Tritium air monitors T-446 and T-290 (flow-through ionization-chamber type with a flow rate of one to two l/min) were used to survey the LLL parking lots and areas along roads north and east of the Laboratory between 0730 and 0830 on the morning of the release. The only positive off-site values (approximately 5 $\mu Ci/m^3$) were recorded with a T-446 monitor a few hundred yards east of the Laboratory boundary. Minimum detectable concentration for this instrument is about 5 $\mu Ci/m^3$.

The samples of airborne water vapor are collected by inserting an aluminum pipe into a dewar filled with liquid nitrogen. When the exposed portion of the pipe becomes cold, atmospheric water vapor condenses on it. After the liquid nitrogen evaporates, the aluminum pipe returns to ambient temperature, causing the ice on the pipe to melt. The melt water (approximately 5 ml) collects in the dewar. A 1-ml aliquot of the sample is then added to the proper liquid scintillant and analyzed for tritium activity in a liquid scintillation counter. The minimum detectable tritium concentration by this method is about 5 nCi/l. If the relative humidity is known, the amount of water vapor per unit volume of air at any particular temperature may be calculated. With this information, and the tritium activity per ml of water, the activity of tritiated water vapor per unit volume of air can be determined. Assuming a relative humidity of 50 percent, the minimum detectable activity for tritiated water vapor is 40 pCi/m^3. The results of this survey, listed completely in Table 1 and summarized in Table 2, are based on a relative humidity of 50 percent. Locations where these samples were taken are shown in Figure 1.

The vegetation samples selected were mostly varieties that remain green during the dry season, so that a reasonable amount of sample water could be obtained for analysis. In general, each sample consisted of a single variety of vegetation and weighed about 250 g as estimated by LLL analytical Laboratory personnel,

Table 1

Airborne water vapor sample tritium activities in pCi/m^3.
Activities lower than 40 pCi/m^3 are not listed.

Sample No.	Series[a]			
	AS-1	AS-2	AS-3	AS-4
1	230	77	1400	260
2	98	---	470	96
3	140	200	310	250
4	200	1200	180	330
5	Lost	650		2600
6	230	1400		630
7	160	880		---
8	120	570		90
9		420		---
10		58		59
11		96		59
12		350		100
13		610		78
14				---
15				170
16				12000
17				---
18				110
19				55
20				9800

[a] Times and locations of samples were as follows:

AS-1: Along State Highway 33 from Interstate 5 through Firebaugh, 1200 to 1700, August 6.
AS-2: On-site at LLL, 0830 to 0930, August 6.
AS-3: On Greenville Road east of LLL, 0800 to 0900, August 6.
AS-4: On-site at LLL, 1350 to 1600, August 6.

Table 2

Summary of airborne water-vapor sampling results

	All samples		Off-site samples		On-site samples	
Activity (nCi/m^3)	No. of samples	Percent of total	No. of samples	Percent of total	No. of samples	Percent of total
<0.1	15	34	1	9	14	42
0.1–0.5	18	41	9	82	9	27
0.5–1	5	11	-	-	5	15
1.0–5	4	9	1	9	3	9
5.0–12[a]	2	5	-	-	2	7
Totals	44		11		33	

[a]Highest value recorded.

Figure 1

Cloud Path and Estimated Arrival Times Based on Meteorological Predictions. Airborne Water-vapor Sample Locations

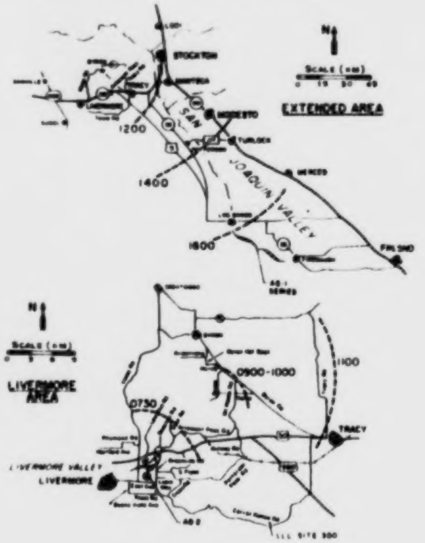

Table 3

Locations where vegetation samples were collected

Sample series	Location
VS-1	Interstate 680 from Danville to Sunol.
VS-2	Altamont Pass Road at Goecken Road.
VS-3	From Byron along Hwy 4 and Tracy Road to Tracy.
VS-4	From Tracy south on Hwy 33 to Patterson, east on J-17 to Turlock, north on Hwy 99 to Manteca, and west on Hwy 120 to Tracy.
VS-5	Vicinity of Lodi.
VS-6	South of Fresno on Hwy 99.
VS-7	Goecken Road.
VS-8	West from Greenville Road along Western Pacific and Southern Pacific RR. tracks.
VS-9	From Hwy 580 on Vasco Road and Camino Diablo Road to Byron, then south on Byron Road and Mountainhouse Road to Altamont Pass Road.
VS-10	From Mountainhouse along Mountainhouse Road, Kelso Road, Bruns Road, Holey Road, Armstrong Road, and Byron Hot Springs Road to Byron.
VS-11	North of Hwy 580 on Laughlin Road, west along Hwy 580 to Vasco Road, north on Vasco Road to Raymond Road, and west on Raymond Road and Hartford Avenue.
VS-12	West on Tesla Road from Las Positas Avenue to Buena Vista Road, north on Buena Vista Road to East Avenue, and intersection of Vasco Road and Western Pacific RR. track.
VS-13	North on Greenville Road from Patterson Pass Road to Hwy 580, West on Hwy 580 to Laughlin Road, north on Laughlin Road.
VS-14	East on Altamont Pass Road from Hwy 580 to Mountainhouse, north on Vasco Road from East Avenue to Hwy 580, east along Hwy 580 to Laughlin Road.
VS-15	North on Dyer Road from Altamont Pass Road.
VS-16	East on Patterson Pass Road from South Flynn Road to Midway.
VS-17	East on Altamont Pass Road from Hwy 580 to Midway Road, south on Midway Road to Midway.
VS-18	Along Flynn Road from Patterson Pass Road to Hwy 580.
VS-19	South on Interstate 5 from Hwy 50 to Hwy 33, east on Hwy 33 through Los Baños to Firebaugh.
VS-20	East on Corral Hollow Road from Cross Road to Hwy 580, west on Hwy 580 to Greenville Road.
VS-21	East on Tesla Road and Corral Hollow from Greenville Road to LLL-Site 300.
VS-22	East perimeter of LLL-Livermore Site.
VS-23	North on Greenville Road from East Avenue to Hwy 580, east on Patterson Pass Road from Flynn Road to Cross Road, south on Cross Road to Tesla Road.
VS-24	East on Lupin Way from Greenville Road.
VS-25	LLL-Livermore Site.

Table 4

Vegetation Sample Tritium Activities in µCi/liter. Activities Lower Than the Minimum Detectable Level of 0.005 µCi/liter Are Not Listed.

Sample No.	Sample Series																									
	VS-1	VS-2	VS-3	VS-4	VS-5	VS-6	VS-7	VS-8	VS-9	VS-10	VS-11	VS-12	VS-13	VS-13 (rerun)	VS-14	VS-14 (rerun)	VS-15	VS-16	VS-17	VS-18	VS-19	VS-20	VS-21	VS-22	VS-23	VS-24	VS-25
1	—	0.009	0.009	—	0.011	—	0.012	0.011	0.006	0.015	0.33	—	—		0.086		0.030	0.009	0.018	—	0.009	—	0.007	0.011	—	0.014	0.064
2	—	0.039	0.009	0.007	—	—	0.007	0.023	0.010	0.017	0.20	0.011	0.011		0.034		0.039	0.007	0.155	—	0.014	—	—	0.023	0.030	—	0.064
3	—	0.027	—	0.009	0.025	—	0.010	0.014	0.015	0.019	0.68	—	—		0.150		0.023	0.009	0.036	0.007	0.009	—	—	0.027	0.014	0.014	0.077
4	—	0.093	0.007	0.007	—	—	—	0.048	—	0.015	0.060	—	—		0.050		0.027	0.009	0.020	0.007	—	—	0.009	0.023	0.018	0.007	0.496
5	—	0.014	—	—	—	—	—	0.035	—	0.010	0.014	—	0.011		0.086		0.007	—	0.110	—	0.009	—	—	0.027	0.023		1.20
6	—		0.007	—	0.009		—	0.025	—	0.024	0.085	—	0.014		0.018		0.007	0.009	0.014	—	—	—	—	0.036	0.014		0.910
7	—		—	0.007				0.210	—	0.028	0.018	—	0.030		0.018		—	—	0.011	0.007	—	0.007	—	0.014	0.016		1.10
8	—		—	—				0.034	—	0.020	0.025	—	0.009		0.018		—	—	—	—	0.014	0.014	—	0.014	0.020		0.170
9	—		—	—				0.020	—	0.015	0.008		0.068	0.090	0.014		—	0.007	—	—	—	0.011	—	0.014	0.018		0.036
10	—		0.007	—				0.080	—	—	0.008		0.68	0.80	0.014		0.011	—	—	—	0.007	—	—	0.023	—		0.043
11	—		—	—				0.091	—	—	0.065		0.018		0.018	0.030	0.007	—	—	—	0.014	0.014	0.007	0.027	—		
12	—		—	—				0.091	—	—	—		0.009		0.830	0.39	0.007	0.007	—	0.014	—	—		0.036	0.011		
13	—		0.007	—				0.320	—	—	0.012		0.300		0.009			—	0.007	0.007	—	0.014		0.055	—		
14	—		0.009	—				0.007	—	0.013	0.007		0.023		0.009			—	—	0.009		--		0.045			
15	—		—	—				0.019	—	—	0.014		0.055		—			—	—	—		0.014		0.007			
16	—		0.007	—				—	0.006	—	0.012		0.007		—			—	0.009			0.009		0.210			
17	—		0.009	—				0.020	—	—	0.007		—		—			—	—			0.009		0.300			
18	—		0.007	—				—	0.025	—	0.012				—			—	0.023			0.023					
19	0.01		0.007	—					0.015	0.007	0.007				—			—									
20			0.014	—						0.005					—			0.007									
21			0.016	—						—					—												
22			0.014	—						0.006					0.007												
23			0.011	—						—					—												
24			0.016	—											—												
25			0.025	—											—												
26			0.021	0.006											—	0.010											
27			0.034	0.015											1.10	0.86											
28			0.018	0.011																							
29				—																							
30 through 42				—																							
43				0.006																							
44				—																							

Table 5

Summary of vegetation sampling results

Activity µCi/liter	All samples No. of samples	All samples Percent of total	Off-site samples No. of samples	Off-site samples Percent of total	On-site samples No. of samples	On-site samples Percent of total
<0.01	250	63	249	68	1	4
0.01–0.05	110	28	95	26	15	56
0.05–0.1	15	4	11	3	4	15
0.1–0.5	12	3	8	2	4	15
0.5–1.2[a]	7	2	4	1	3	10
Totals	394		367		27	

[a] Highest value recorded.

but this value varied considerably.

Water was extracted from the vegetation by a freeze-drying technique [1]. A small portion of the original sample was placed in a vacuum flask and frozen by immersing the flask in liquid nitrogen. This fixed the water to the vegetation during the evacuation process. A 50-µ vacuum was pulled on the sample and the evolved water was collected in a liquid nitrogen cooled receiver, while the sample flask returned to room temperature. The water-vapor transfer was allowed to proceed until a minimum of 1 ml of water was collected in the receiver. Tritium activity in the collected water was measured with a liquid-scintillation counter.

Table 3 lists the locations where vegetation samples were collected. The landmarks used for identification may be found in Figure 1. Table 4 lists the tritium activity of these vegetation samples, and Table 5 summarizes the vegetation-sampling results.

Water samples were collected from storage ponds and tanks in one-liter plastic bottles at the locations shown in Figure 2. One ml of each sample was analyzed with a liquid-scintillation counter. Only six locations indicated levels above 5 nCi/l. These were WS-6, WS-20, WS-21, WS-22, WS-24 and WS-26, with activities ranging from 7 to 14 nCi/l. Milk samples were collected at a number of locations in the Livermore and San Joaquin Valleys. These samples were treated by a freeze-drying process similar to that used for treating the vegetation samples. Figure 2 shows the locations where these samples were collected. Only three samples indicated activities above the 5 nCi/l detection limit. Samples MS-3, MS-5 and MS-13 showed tritium activities between 6 and 8 nCi/l.

Based on release parameters, including meteorological conditions, and analysis of environmental samples, it was established that the potential exposure risk to individuals both on-site and off-site was very low. However, urine samples were collected to confirm these predictions. These urine samples were prepared for analysis by distilling off the water and counting 1 ml of the distillate in a liquid-scintillation counter.

Figure 2

Water and Milk Sample Locations

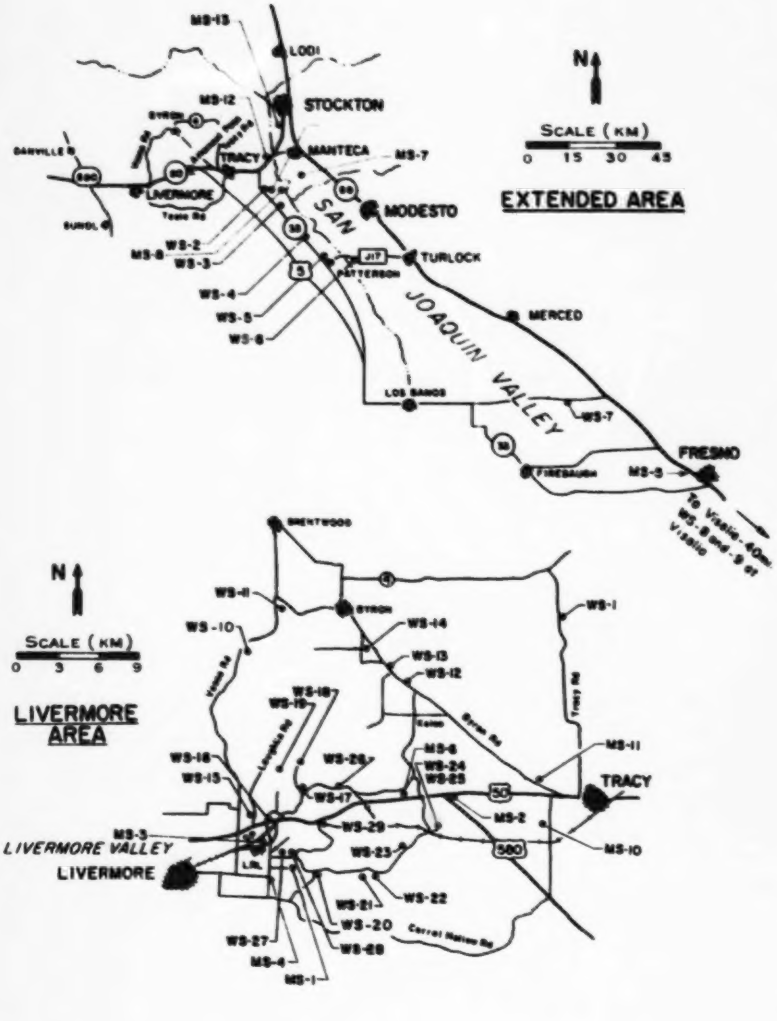

Table 6

Comparison of tritium activities for samples taken before and after accidental tritium release. Values are in nCi/l

Sample	Max-accident levels	Pre-accident levels	Off-site MPC	Detection limit
Water	14	<5	3000 b	5
Air (Water vapor)	$1.4X10^{-3}$ a	$<4X10^{-5}$ a	$2X10^{-1}$ a	$4X10^{-5}$ a
Milk	8.0	<5	3000 b	5
Vegetation (water)	1000	10(av) 250(max)	3000 b	5
Urine	<5	<5	3000 b	5

[a] The minimum sensitivity would depend on the relative humidity, which was 50 percent at the time of the release.

[b] Assuming that tritium was equilibrated in the environment. This would be true only for a chronic release situation, but the values are useful as reference points.

Urine samples were obtained from 31 LLL employees and 19 other individuals who were in the area to the north and northeast of the Laboratory along Greenville and Altamont Pass Roads at the time of cloud passage. No activity above the minimum detectable level of 5 nCi/l was found. Urine samples were also collected from two cows quartered in the LLL Bio-Medical Division Facility on August 6, 1970. This facility is located 1.3 km north of the exhaust stack of the Gaseous Chemistry Building. The activities measured were 28 and 45 nCi/l, respectively.

Accidental Release Levels as Related to Maximum Permissible Concentrations

Table 6 provides a comparison between the levels of off-site tritium activity measured after the release with previous measurements (which established background levels), off-site MPC, and the minimum measurable activities for the listed samples. All environmental samples of water, milk and urine contained normal background levels of tritium. Tritium was detected in the vegetation and water vapor in the air; however, even the highest concentrations were below off-site MPC for continuous exposure. The maximum values obtained from on-site samples were 12 nCi/m^3 for water vapor and 1.2 μCi/l for vegetation. The on-site MPC are 100 μCi/l for vegetation, assuming equilibration in the environment, and 5 $\mu Ci/m^3$ for water vapor.

Hazards Analysis

There are two potential areas of concern regarding exposure to individuals off-site as a result of the release. They are 1) the dose an individual could have received had he been located downwind at the point of maximum ground concentration during the entire passage of the cloud, and 2) the dose an individual could have received through the forage-cow-milk exposure pathway from the tritium deposited on vegetation.

Given the meteorological conditions that existed at the time of release (very stable with light winds) and the stack height (36.5m) the estimated point of maximum ground-level concentration is about 1 to 1.2 km downwind. This distance coincides approximately with the nearest off-site location in the downwind direction. If it is assumed that an individual were standing on the centerline of the cloud's track at the nearest downwind off-site location and that one percent of the tritium was present in the more hazardous oxide form, the estimated dose to this individual would be about 3 mrem. As a point of reference, if we assume that all of the tritium was converted to the oxide form (which would be likely only in the event of a fire or explosion at the point of release), the maximum projected exposure at the site boundary would be 300 mrem. This compares with a maximum permissible annual dose to a nonoccupationally exposed individual of 500 mrem. Since all urine samples collected off-site indicated that there was no tritium present above detectable levels (5 nCi/l), the maximum undetectable dose which could have been received from exposure to the cloud would have been about 0.025 mrem.

The second potential concern is the dose that could be received by an individual drinking milk produced by cows that have grazed on contaminated vegetation. As noted earlier, positive indications of tritium were found in off-site vegetation, with the maximum being 1.0 μCi/l in one sample. However, there was no indication of tritium in water or milk above normal levels.

If we make the conservative assumption that a large area of pastureland was uniformly contaminated to the maximum level observed, the dose to a child drinking the milk from cows grazing on this vegetation can be estimated by techniques used to predict internal dose from radionuclides released to the biosphere [2]. If we assume that there were about four liters of water per square meter of vegetation, then the concentration at the point of maximum deposition would be $(4\ l/m^2) \times (1.0\ \mu Ci/l) = 4.0\ \mu Ci/m^2$. This level would result in a dose of 70 mrem to a child exposed via this pathway. This value should be considered a conservative maximum because it assumes large-area contamination at the highest level found at one location in the survey, which is actually unlikely because a) no other locations reaching this level of contamination were found and b) it is unlikely that any such locations were missed because of the extensiveness of the survey. This conservatism is further supported by the fact that no tritium activity above background levels was found in any of the milk or urine samples collected.

Conclusions

The results from this environmental and bioassay sampling program indicate that no significant exposure to on- or off-site individuals resulted from the release. All samples of water, milk, and urine contained normal background levels of tritium. Although detectable levels of activity were found in some vegetation and atmospheric water vapor samples, the highest measured concentrations as well as the calculated minimum credible ground-level air concentrations during cloud passage were well below the off-site maximum permissible concentrations for continuous exposure. If all of the tritium was assumed to be converted to the oxide form, the calculated maximum exposure at the site boundary as a result of exposure to the cloud would be 300 mrem. Since tritium was not present in urine samples above detectable levels, the maximum undetectable dose which could have been received would have been about 0.025 mrem. Based on vegetation-sampling results, the maximum credible dose to a child exposed via the forage-cow-milk pathway would be about 70 mrem.

ACKNOWLEDGMENTS

The authors acknowledge the assistance of the many members of the Hazards Control Department who participated in the initial emergency response and the extensive sample collection, preparation and counting programs. In addition, special thanks go to Todd Crawford and Joe Knox from the Plowshare Division who supplied early meteorological information used to predict the path of the cloud during the first several hours after release, Amasa Cornish of the State Department of Public Health who assisted in the collection of milk and urine samples, the Bio-Medical Division, which assisted in the collection of on-site vegetation samples, and R. G. Barton of the Technical Information Department for his advice and assistance in preparing these data for publication.

This work was performed under the auspices of the U.S. Atomic Energy Commission.

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ENVIRONMENTAL TRITIUM STUDIES AT A PWR POWER PLANT

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Tritium produced by natural or artificial processes in the atmosphere is oxidized to form water, and follows the hydrologic cycle. Before the introduction of large quantities by thermonuclear detonations, levels of tritium in the environment were very low and were studied only as a scientific curiosity. Because of the extremely low concentrations of tritium even after the advent of bomb tests and the resulting measurement problems, the retention or incorporation characteristics of tritium in biota and sediment in the organically bound state are not well known. With the expansion of the nuclear power industry, increased quantities of tritium are expected to be released in localized areas, and there is a need to know more about the behavior of tritium in the environment.

This study was designed to define the relative concentrations of tritium in several compartments of an estuarine environment. Measurements were made of tritium in the loose and organically bound water in bottom sediments, rooted aquatic plants, fish, and ambient water.

The area selected for this study was a thirty mile section of the Hudson River estuary which includes the nuclear power facility at Indian Point, Figure 1. This pressurized water reactor has been in operation since 1962 and is presently releasing 400 to 1100 curies of tritium per year to the estuary [1]. In this region, it is possible to obtain current background concentrations in the river from fallout, by collecting samples upstream of the reactor beyond the range of upstream transport, and to determine whether the tritium from the nuclear power plant can be detected in the environment above the present ambient levels.

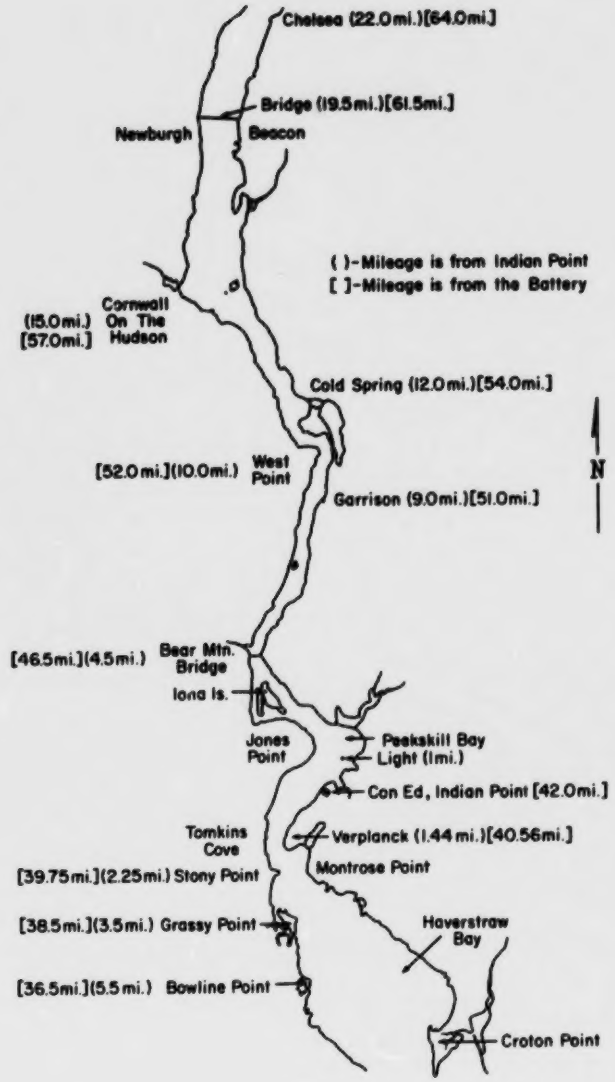
Sample Collection and Preparation

A set of four continuous water samplers were set up along the river. One sampler was placed at Chelsea, New York, at the northern end of the section studied, to monitor background levels in the river. Samplers were set up at Verplanck and Iona Island, 1.5 miles downstream and 4.5 miles upstream from the discharge point, respectively, to determine whether additional tritium from the plant could be detected in the water. To monitor the effluent from the plant, a fourth sampler was set up to collect samples in the discharge canal. This network was supplemented by one-liter samples of water collected monthly at eight to twelve locations to determine the spatial variations in this region.

**Present Address: NUS Corp., Rockville, MD*

Figure 1

Hudson River



Chelsea (22.0mi.)[64.0mi.]
Bridge (19.5mi.)[61.5mi.]
Newburgh
Beacon
( )-Mileage is from Indian Point
[ ]-Mileage is from the Battery
(15.0mi.) [57.0mi.] Cornwall On The Hudson
Cold Spring (12.0mi.)[54.0mi.]
N
[52.0mi.](10.0mi.) West Point
Garrison (9.0mi.)[51.0mi.]
[46.5mi.](4.5mi.) Bear Mtn. Bridge
Iona Is.
Jones Point
Peekskill Bay
Light (1mi.)
Con Ed, Indian Point [42.0mi.]
Tomkins Cove
Verplanck (1.44 mi.)[40.56mi.]
[39.75mi.](2.25mi.) Stony Point
Montrose Point
Haverstraw Bay
[38.5mi.](3.5mi.) Grassy Point
[36.5mi.](5.5mi.) Bowline Point
Croton Point

A species of pondweed, *Potamogeton perfoliatus*, indigenous to this region was chosen as representative of the rooted aquatic vegetation. Samples weighing 1 kg wet were taken biweekly through the entire growing season at one site to observe seasonal patterns in the tritium uptake. In the summer, samples of this species of vegetation were collected at locations covering the entire section. Samples of bottom sediment were obtained with an Emory dredge sampler at the same time grab water samples were being taken. Only the undisturbed portion of the sample was collected for analysis. The total sample of sediment weighed approximately 1 kg (wet).

Fish for the study were caught near the plant, and were supplemented by fish bought from commercial fishermen fishing downstream from the plant. No preservatives were used for any sample, all fish samples were frozen while still fresh.

The raw water samples were filtered through a 0.45 μm millipore filter to remove suspended materials. The samples were distilled to remove remaining impurities and were electrolytically enriched before counting. Electrolytic enrichment of water samples was accomplished in a system developed in this laboratory [2]. Fifty ml of water was introduced into an electrolysis cell with two coiled platinum electrodes at the bottom of the cell which are completely immersed in the sample at all times. An indentation at the bottom of the cell permits excellent cooling of the samples through high surface area contact, and also helps to hold the electrodes in position. The sample was acidified with sulfuric acid. This served the dual purpose of acting as the electrolyte for current conduction and providing a means of measuring the volume change by determination of initial and final acid concentrations.

Seven cells were connected in series and placed in a cooling bath maintained at $8^{\circ}C$. Electrolysis was performed at a constant current of 3A resulting in a current density of $200\ mA/cm^2$. One known tritium solution was included with the samples in each run for standardization and quality control. From the enrichment and volume change of the known solution, the enrichment of the other samples was calculated using the measured volume change determined for each sample. With known tritium solutions in each cell, the reproducibility of enrichment between the seven cells was found to be within ±4% (1σ) of the mean value for a single enrichment run.

After electrolysis, the samples were distilled to remove the sulfuric acid. Five ml of sample were added to 20 ml of cocktail (Aquasol - New England Nuclear) and counted four times for 100 minutes each, a total 400 minutes counting time. Background blanks, standards and known tritium solutions used in the laboratory were calibrated with a tritiated water standard certified and supplied by New England Nuclear. In this counting system, the background is 8.50 cpm, and the efficiency is 16%. The minimum detectable activity for this system is 100 pCi/l.

It was found that freeze-drying was the most convenient method to collect loose water and was the least likely to produce any unwanted change in the fish, vegetation, and bottom sediment samples. After drying, the sample was compressed to produce a more even-burning reaction during combustion. The size of the dried sample usually ranged from 100 g for vegetation to 250 g for fish or sediment samples.

The combustion system based on the design by Drobinski, *et al.* [3], consisted of a series of four stainless steel tube furnaces each fitted with heaters and thermocouples to maintain the proper temperatures. The units were connected with stainless steel tubing, connectors and valves capable of withstanding high temperatures and corrosive atmospheres. Pressure and vacuum gauges and flow meters were installed throughout the system to meter the flow and monitor the conditions during operation. Oxygen and argon were supplied from tanks through regulators to provide the proper oxidizing atmosphere. Water was collected in a trap immersed in an isopropanol-dry ice bath. Once the sample was sealed in the main furnace, the temperature was raised to 800°C. The oxidized and partially oxidized gases flowed through the furnace to two furnaces packed with CuO (600°C) and SiO_2 (45°C) where the oxidation of the hydrogen and carbon monoxide was completed. A fourth furnace, filled with copper wool, silver wool and manganese dioxide at 200°C, removed oxides of nitrogen, sulfur and halogens from the hot gases. The water collected in the trap was then treated as other water samples. The efficiency of the system to convert hydrogen in organic material to water was over 92%. This was determined by burning glucose.

Results and Discussion

Seasonal variation of tritium levels in the river can be seen in Figure 2. These values for continuous samples collected at two-week intervals at Chelsea represent background concentrations from fallout. The concentrations ranged from 400 to 800 pCi/l depending on the season of the year. In 1970, the average concentration in the river was 580 pCi/l.

Tritium concentrations in surface waters in the United States have sharply decreased since they reached a peak in the mid-1960s [4]. This trend was also observed in the Hudson River as shown in Figure 3 [2,4-7]. In the last few years, tritium levels in surface waters such as the Potomac and the Susquehanna Rivers have declined at a much slower rate than before. To determine whether there was a significant decrease in tritium concentration in the Hudson River, over the study period, the average tritium concentration in an eight-month period from November 1969 to June 1970 was compared to a similar period from November 1970 to June 1971. It was found that the difference in average tritium concentrations for these two periods was less than five percent. With the many variables affecting the tritium levels in the water, the magnitude of this is not meaningful. It is clear, however, that the trend in tritium concentration in the Hudson River closely parallels those of the Potomac and Susquehanna Rivers.

The annual amount of tritium discharged into the river from the reactor at Indian Point is small compared to the estimated 200 kCi of tritium deposited into the Hudson River Estuary System each year. This estimate was calculated by multiplying the average rainfall on the basin (122 cm) by the area of the river basin (3.4×10^9 m^2) and the average tritium concentration in precipitation (500 pCi/l). However, during the year when

Figure 2

Background Tritium Concentration in Water at Chelsea, New York - 22.0 Miles North of Indian Point (64.0 Miles North of the Battery)

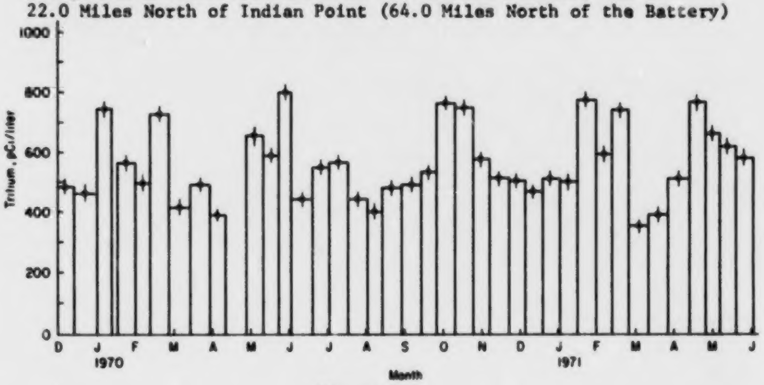
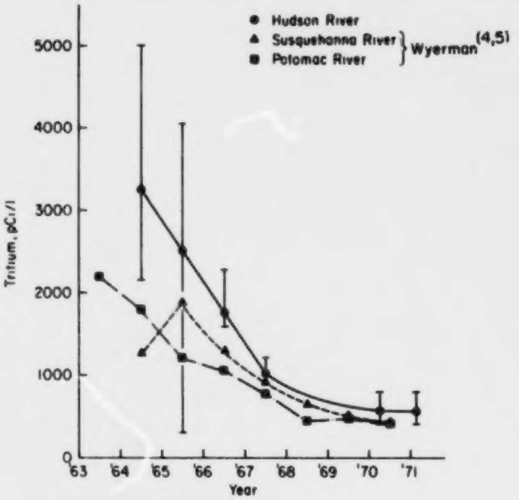


Figure 3

Average Tritium Concentrations, in pCi/liter, in the Hudson River, the Susquehanna River and the Potomac River



Hudson River Data

1964 } Chesnutt(6)
1965 }
1966 Cosolito(2)
1967 Cosolito(7)

Figure 4

Tritium Concentration in Water at Verplanck Station - 1.5 Miles South of Indian Point (40.5 Miles North of Battery) Compared to Chelsea, New York (64.0 Miles North of Battery)

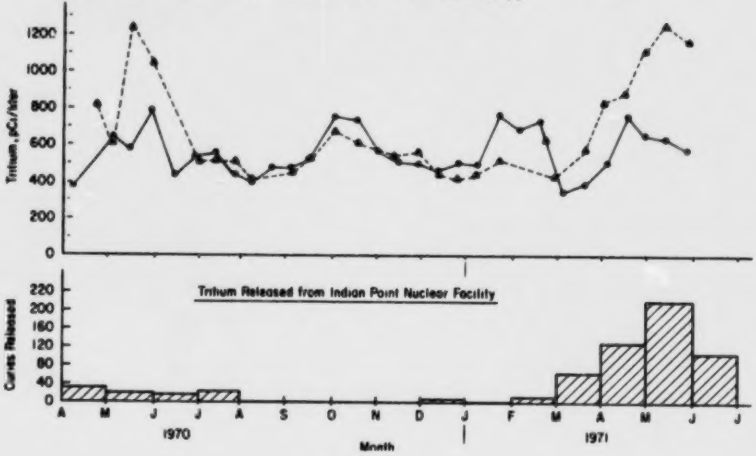


Table 1

Tritium Concentrations in Grab Water Samples

(pCi/1 ±1σ)

Location	Miles from Battery	April 1970	May 1970	June 1970	July 1970
Hyde Park	76.0	400 ± 30	560 ± 60	530 ± 40	560 ± 30
Newburgh	61.5		550 ± 70	560 ± 40	530 ± 30
Cold Spring	55.5				560 ± 30
Bear Mt.	46.5	400 ± 30	530 ± 40	440 ± 30	530 ± 30
Iona Island	45.0	420 ± 30	630 ± 40		420 ± 20
Peekskill Bay	43.0	460 ± 40	630 ± 60	530 ± 30	450 ± 30
Con Ed Discharge Point	42.0	470 ± 30	650 ± 30	660 ± 40	460 ± 30
Con Ed Mid Channel	42.0	370 ± 30	500 ± 30	530 ± 30	540 ± 30
Con Ed West Side	42.0	660 ± 40	650 ± 70	560 ± 40	450 ± 30
Tompkins Cove	41.0		560 ± 30	500 ± 40	500 ± 40
Verplanck	40.5	440 ± 40	580 ± 50	630 ± 30	670 ± 30
Stony Point	39.5	430 ± 30	810 ± 30	470 ± 40	
Montrose Point	39.5		830 ± 50		620 ± 30
Bowline Point	36.0		490 ± 60	600 ± 40	570 ± 60
Croton Point	35.0				

Location	Miles from Battery	August 1970	September 1970	October 1970	November 1970
Hyde Park	76.0				
Newburgh	61.5	575 ± 30	525 ± 25	560 ± 30	
Cold Spring	55.5	500 ± 25	490 ± 30	550 ± 30	
Bear Mt.	46.5	480 ± 25	520 ± 30	460 ± 30	
Iona Island	45.0	750 ± 25	680 ± 30	390 ± 40	620 ± 30
Peekskill Bay	43.0	510 ± 25	470 ± 30	470 ± 30	650 ± 30
Con Ed Discharge Point	42.0	610 ± 40	675 ± 30	490 ± 30	630 ± 30
Con Ed Mid Channel	42.0	560 ± 40	470 ± 30	580 ± 30 530 ± 40	620 ± 30
Con Ed West Side	42.0	520 ± 40	470 + 30	670 ± 30	530 ± 30
Tompkins Cove	41.0	480 ± 40	630 + 40	630 ± 40	560 ± 30
Verplanck	40.5	600 ± 20	530 + 40	390 ± 40	700 ± 30
Stony Point	39.5	530 ± 40	520 + 30	660 ± 30	650 ± 30
Montrose Point	39.5		490 + 40		600 ± 30
Bowline Point	36.0	420 ± 20	480 + 30	450 ± 40	
Croton Point	35.0	500 ± 20	490 + 30		620 ± 30

the reactor discharges tens of curies of tritium per month, it is possible to detect tritium in the water above the ambient levels. This was seen at Verplanck during the spring of 1970 and 1971, when the concentrations in the water were a factor of two higher than what was observed in upstream samples, Figure 4. In the summer and fall of 1970, when essentially no tritium effluent was discharged from the plant, the concentrations were in agreement with the background values.

The tritium concentrations measured at Iona Island sampling station, 4.5 miles north of the reactor, were compared to the concentrations measured at Chelsea. The average concentration at Iona was 535 pCi/l. This was nine percent less on the average than that measured at Chelsea for the same time period. This small variation may be the result of differences in sampling techniques used in the two locations.

Grab water samples collected monthly in the river are listed in Table 1. No systematic variations are observed. The results were quite variable, which can be attributed to the precision of the measurements and physical processes occurring in the river, such as vertical mixing, exchange at the water-air interface and the introduction of ground water or local runoff. It would be difficult at the present time to monitor the sporadic discharge of effluent from the reactor into the surrounding water using grab water samples without knowing the mixing characteristics and currents in this section of the river and the time of release of the effluent.

The tritium concentrations measured in the Hudson River during 1970 were compared to tritium measurements of New York City tap water originating in the same watershed area [8] and with other tritium measurements of surface waters in the northeast [5]. The results between the three sources were in good agreement.

Sediments

Bottom sediments play an important role in the movement and retention of radionuclides in the aquatic environment. The exchange and retention capabilities of sediments for radionuclides has been shown to be related to the organic content and the exposed hydroxyl groups of the clay minerals in the sediments [9]. The behavior and interaction of tritiated water with bottom sediments has not been studied, especially under environmental conditions. In laboratory studies under static conditions, however, Stewart [10] has demonstrated that isotopic exchange and fractionation can take place when clays are saturated with tritiated water for long periods of time, producing bound tritium concentrations two to four times greater than in the aqueous phase.

In the Hudson River, bottom sediments contain organics in the range of six percent by weight [9]. This organic fraction is composed of decayed plant and animal life that has accumulated over the years when tritium concentrations in water were as much as four to eight times higher than present levels, as was the case in 1964, Figure 4. Samples of bottom sediments were taken to determine the extent to which tritium is incorporated by the aforementioned exchange and organic mechanisms. If the residence time of tritium in bottom sediments is long compared to time in

Table 2

Tritium Concentrations in Sediments

December, 1969

Location	Miles From Battery	Ambient Water pCi/l	Loose pCi/l H_2O	Bound pCi/l H_2O
North Inlet Con Ed	42.0	470 ± 30	530 ± 80	5,200 ± 160
Con Ed Mid Channel	42.0	600 ± 30	720 ± 30	12,200 ± 250
Con Ed West Side	42.0	340 ± 40	540 ± 40	3,800 ± 270
Average		470	600	7,000

Table 3

Tritium Concentrations in Sediments

April, 1970

Location	Miles from Battery	Ambient Water pCi/l	Loose pCi/l H_2O	Bound pCi/l H_2O
Hyde Park	76.0	400 ± 30	640 ± 25	1500 ± 120
Bear Mt.	46.5	400 ± 30	430 ± 40	4200 ± 110
Iona Island	45.0	420 ± 35	430 ± 40	5700 ± 120
Peekskill Bay	43.0	460 ± 35	1100 ± 50	2400 ± 125
Con Ed Canal	42.0	470 ± 30	970 ± 50	8100 ± 155
Con Ed Mid Channel	42.0	570 ± 30	600 ± 40	6300 ± 150
Con Ed West Side	42.0	660 ± 40	500 ± 20	1300 ± 120
Verplanck	40.5	440 ± 40	490 ± 20	2900 ± 120
Stony Point	39.5	430 ± 30	600 ± 30	6900 ± 150
Average		470	690	4400

Table 4

Tritium Concentrations in Sediments

August, 1970

Location	Miles from Battery	Ambient Water pCi/l	Loose pCi/l H_2O	Bound pCi/l H_2O
Newburgh	61.5	570 ± 30	890 ± 35	1050 ± 115
Cold Springs	55.5	500 ± 25	940 ± 35	340 ± 110
Iona Island	45.0	490 ± 25	N.S.	5900 ± 140
Peekskill Bay	43.0	700 ± 25	1060 ± 30	770 ± 110
Con Ed Canal	42.0	600 ± 40	1300 ± 30	7100 ± 150
Con Ed Mid Channel	42.0	540 ± 40	1300 ± 30	5600 ± 150
Con Ed West	42.0	520 ± 40	1060 ± 30	540 ± 120
Verplanck	40.5	500 ± 20	820 ± 30	8500 ± 160
Stony Point	39.5	530 ± 40	1000 ± 30	8340 ± 165
Buoy #14	36.0	420 ± 20	1000 ± 30	17800 ± 200
Average		530	1040	5600

Figure 5

Bound Tritium Concentration in Bottom Sediment vs. Miles - August 1970

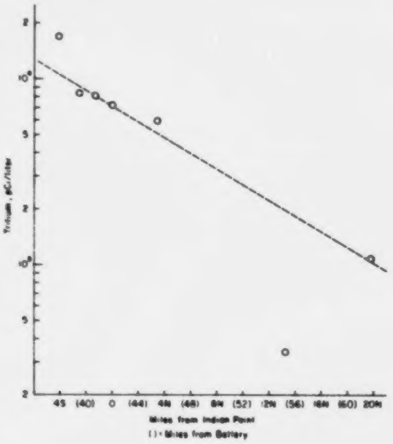
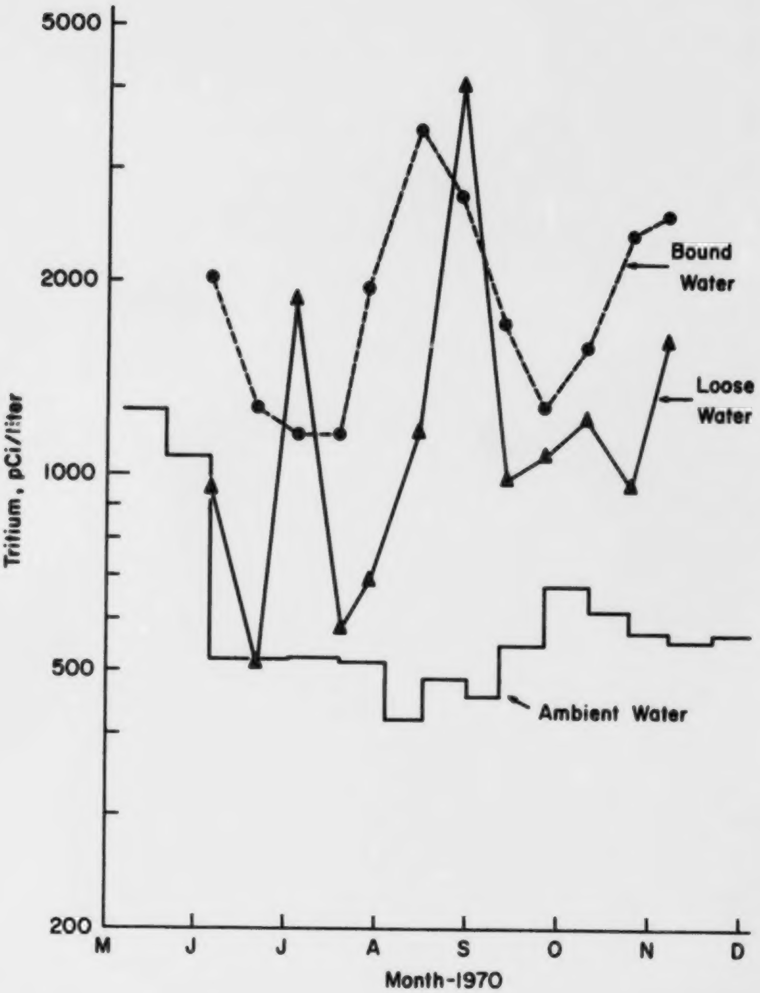


Figure 6

Tritium Concentrations in Potamogeton Perfoliatus at Vernlanck Station, Mile 1.44 South (Mile 40.5)



plants and fish, the sediments might act as a sink or reservoir for tritium in the river.

The results for sediment samples are shown in Tables 2 to 4. The loose water component of bottom sediment is defined as the water removed by freeze drying. This would include interstitial water, capillary water and some water of hydration. The average tritium activities in the loose water were 600 pCi/l and 570 pCi/l in December and April, respectively, see Tables 2 and 3. This was 15 to 20% higher than ambient water collected at the same time. These differences are significant at the 95% confidence level as shown by the paired t test. The tritium activity in the loose water recovered from the samples collected in August is shown in Table 4. In these samples, tritium in loose water was two times higher than the ambient water. This is, of course, a highly significant difference. This phenomenon was also observed in fish and plant samples described later in this paper.

Bound tritium in bottom sediments was recovered in the form of water by heating dried sediments to 800°C in an oxygen atmosphere. The specific activity of the bound tritium expressed in terms of pCi/l water varied from 540 to 17,800 pCi/l with an average of 5,600 pCi/l. This was a factor of ten greater in concentration than ambient water. The range of the specific activities between samples at the different locations was about one order of magnitude. This variability may be due, in part, to the sampling sites. The physical characteristics of the location, such as average depth, currents and physical obstructions, will change the flow rates and consequently the sedimentation rates. This would alter the composition and size fractions of the sediment, which may account for the change of properties.

In the downstream portion of the region, the cross-sectional area of the river increases, reducing the average flow rate and increasing the salinity of the water. These two factors result in the flocculation and deposition of suspended sediment of finer size fractions. As might be expected, with the higher exchange capacity of the finer particles of clay and silt, the tritium concentrations in the more southerly sediments will be higher. Bound tritium in bottom sediments does increase with distance downstream. This effect is shown in Figure 5.

It was found that sediment samples collected at the Indian Point discharge canal were higher in bound tritium than those from locations directly across the river on some occasions. This suggests that the effluent from the reactor can influence the bound tritium concentrations in the sediment. Through the dispersal processes in this estuary, although these are not well understood, sediment from the reactor outlet could be distributed up and down the river, following the deposition patterns of the sediments. It is possible, then, that this is another mechanism which could enhance or produce the higher bound tritium concentrations in downstream samples as shown in Figure 6.

Plants

Potamogeton perfoliatus, pondweed, is a rooted aquatic plant indigenous in this section of the river. It appears in the river in late spring and it can be found until the end of November. Pondweed flourishes in protected areas, such as shallow coves and bays near the shore. During the growing season, the underground stems embedded in the sediments produce a large number of branched leafy stems. In the winter, the stems decay leaving terminal buds and rhizomes to propagate the plants in the spring. Water and nutrients are taken into the plant through the roots, stems and leaves [11].

The seasonal variation in the tritium concentration in the loose water fraction and organically bound component of Potamogeton was studied at Verplanck throughout the growing season. The bound tritium converted to water was expressed in terms of pCi/l to simplify the comparison with the water fractions. Bound tritium activity showed a buildup in the late summer and early fall, peaking in the middle of September, Figure 6. This accumulation occurred at the time of year when the fresh water flow was at a minimum and the salt water intrusion moved upstream. As the fresh water flow increased, the activity in the tissue appeared to decrease. The activity in this component varied from 1,200 to a high of 3,500 pCi/l. During the growing season (May-November), the average value was 2,000 pCi/l. On an activity basis, the bound tritium in pCi/l was three times higher than the ambient water.

For aquatic plants immersed in water, the loose water fraction of the plant was defined as the water collected from freeze-drying after air-drying the plant for 24 hours at room temperature. Air-drying will remove the bulk of the surface water which would mask any difference from the intracellular and extracellular water in the plant. The activity in loose water increased at the beginning of July during the plants' maximum growth period and once again at the end of August following the buildup of bound tritium.

The tritium activity in the loose water varied from a low of 520 pCi/l, the ambient water concentration, to a high of 4,000 pCi/l, with an average concentration of 1,380 pCi/l. This was a factor of two above the ambient water. The tritium activity in the loose water of the plants increased as the tritium activity in loose water of sediment increased. This suggests a possible intake route for tritium which would enable the plant to accumulate tritium above ambient water concentrations.

It has been found by Cohen [12] that rooted aquatic plants exposed to tritiated water alone would not accumulate tissue bound tritium above the ambient water concentration in laboratory studies. In these studies, the sediment was not labeled with tritium. This was similar to the results of Weinberger and Porter [13] for chlorella. In the river, however, the rooted plants grew from sediment through terminal buds and were exposed to tritium higher than the ambient water. Throughout the season, the plants could obtain additional tritium through the root-sediment contact, which enables the plant to accumulate bound tritium in excess of ambient water concentrations.

At the end of July, a series of eighteen samples of Potamogeton were collected from various locations along the river. Data from the analysis of these plants are presented in Table 5. The pattern found at Verplanck was also observed in these samples. The tritium concentration in the loose water of these samples ranges

Table 5

Tritium Concentrations in *Potamogeton* in Hudson River

July, 1970

Location	Mileage	Ambient Water pCi/l	Loose pCi/l H_2O	Bound pCi/l H_2O	Total Plant pCi/l	Relative Concentration
Cold Spring	55.0	530 ± 30	2600 ± 40	3300 ± 120	2300	4.33
Garrison	52.0		980 ± 30	1800 ± 100	970	1.96
Con Hook	49.0		650 ± 30	1200 ± 120	650	1.31
Manitou	47.8		770 ± 30	1120 ± 110	790	1.60
Iona Island N.	46.0	530 ± 30	1070 ± 40	6300 ± 140	1400	2.66
Iona Island S.	44.8	530 ± 30	1600 ± 115	1420 ± 110	2700	5.50
Fish Island	44.5		730 ± 40	1140 ± 100	730	1.67
Peekskill Bay N.	43.5		980 ± 30	1300 ± 140	920	1.85
Peekskill Bay S.	43.0	430 ± 20	1200 ± 35	1150 ± 110	1100	2.56
Tompkins Cove	41.0	450 ± 30	730 ± 30	1100 ± 100	700	1.56
Verplanck	40.5	520 ± 40	700 ± 30	2000 ± 120	1250	2.21
Stony Point	39.5	470 ± 33	730 ± 30	1400 ± 120	730	1.56
Furnace Brook	39.0		740 ± 40	1240 ± 110	720	1.65
Grassy Point	38.5	420 ± 30	650 ± 30	2300 ± 120	730	1.73
Haverstraw	37.5		700 ± 30	1480 ± 120	720	1.44
South Haverstraw	36.5		960 ± 40	1440 ± 120	920	1.85
Croton-Hudson	36.0		1500 ± 40	1930 ± 110	1600	2.84
Average		490	990	1900		2.08

Table 6

Tritium Concentrations in Hudson River Fish

Species	Collection Date	Loose pCi/l H_2O	Bound pCi/l H_2O	Relative Concentration
White Perch	1/70	600 ± 30	5000 ± 180	1.3
	10/70	500 ± 30	1700 ± 150	0.7
	11/70	660 ± 30	2300 ± 140	1.0
	12/70	870 ± 35	1400 ± 120	1.4
Striped Bass	5/70	1000 ± 50	1700 ± 120	1.0
Alewife	5/70	1050 ± 30	4300 ± 210	1.3
	11/70	800 ± 30	2800 ± 140	1.6
Shad	4/70	560 ± 30	1100 ± 120	1.1
	5/70	760 ± 40	1300 ± 110	1.0
	5/70	960 ± 30	2200 ± 270	1.0
Catfish	6/70	1800 ± 50	1200 ± 110	1.4
Eel	5/70	1400 ± 30	6900 ± 170	2.1
	8/70	890 ± 30	10200 ± 150	3.8
	10/70	590 ± 30	1400 ± 150	1.0
Carp	6/70	1440 ± 45	1800 ± 130	1.0
Tomcod	8/70	470 ± 20	4300 ± 460	1.8
	12/70	1260 ± 30	9400 ± 160	2.0
Average		890	3500	1.5

from 650 to 2,370 pCi/l with a mean value of 990 pCi/l, two times higher than ambient water. The bound tritium concentration varied from 1,140 to 6,300 pCi/l, with a mean value of 1,900 pCi/l. No distinct spatial pattern was seen in these plant samples.

Rooted aquatic plants will incorporate tritium from the water or from the sediment by exchange or metabolic processes. As seen in Figure 6, the concentrations in the plant will vary throughout the growing season. The amount of tritium taken up by the plants may depend on the concentration of tritium in the water, the availability of tritium in the sediment and the growing stage of the plant. In the summer, when these plants are in the maximum growth period, water and ions taken in through the leaves and roots are metabolized at a maximum rate. This will produce the accumulation of tritium in the tissue of the plant observed in late summer. Although the tritium concentrations in the plants are not constant, it is useful for evaluation of the hazard of tritium to calculate the relative concentration of tritium in rooted aquatic plants. In these plants, 86% of the plant is water and 14% of the plant is solid material. Water of combustion accounts for seven percent of the total weight of the plant. From the free and bound water contents and the tritium concentration, a relative concentration of 2.0 was calculated.

Fish

Fish are an important component of the aquatic environment. They serve as a direct link for tritium in the environment to reach man. In the Hudson River estuary, there are numerous varieties of fish. The fish found at any particular location will depend on the salinity of the water, time of the year, migration and feeding habits, and the method of collection. Fish analyzed in this study were species abundant in this section of the river, and they were to some extent eaten by local inhabitants. They included white perch, striped bass, tomcod, carp, shad, eels and catfish. The tritium analysis of the fish is presented in Table 6. The fish taken were small in size and less than one year old with the exception of carp, shad and striped bass. The larger size and weight of these fish suggested that they were two to four years old.

In fresh fish, the loose water fraction accounts for 75% of the wet weight of the fish. Loose water was defined as the intracellular and extracellular water removed by freeze-drying. In this component, the tritium concentration varied from 465 to 1,820 pCi/l, with an average concentration of 880 pCi/l. The bound tritium concentration expressed in terms of pCi/l H_2O ranged from 1,180 to 10,250 pCi/l. The average specific activity in the dried fish was 3,460 pCi/l. The specific activity of the bound tritium, therefore, was four times higher than the loose water. This compared favorably with the ratio of tritium in water of combustion to loose water of 5.6 as reported by Bogen, _et al._ [14] for fish. The dried fish represents only 25% of the total weight and 12% of the hydrogen concentration of the total wet weight of the fish. If the tritium concentrations in pCi/l in both fractions are weighted by the percent of the total weight, a tritium concentration for the wet fish can be determined.

Using the tritium concentration of ambient water and the above factors, a relative concentration of 1.5 was computed for fish.

Bound tritium concentrations varied in different species of fish. The young fish, tomcod and eels were observed to have the highest concentration of bound tritium, six to eight times higher than tritium in loose water. Alewives and white perch had bound tritium concentrations approximately four times higher than loose water. Large and older fish such as shad, carp and striped bass were found to have bound tritium concentrations two times higher than in the loose water.

The interpretation of this relative concentration for tritium and the high bound tritium loose water ratio must be done with care. In an environmental study, the exposure of fish to the ambient environment is not controlled. Although the ambient water levels have been measured and defined, fish, unlike rooted plants, are mobile and able to change their environment. Shad, for instance is a migratory fish. It lives on the ocean floor most of its life and returns to the estuary to spawn. Thus, exposure to tritium from the water and food will vary. The feeding habits of these fish will change with location, time of the year, abundance of vegetation and other factors. The analysis of the stomach content of some species of Hudson River fish presents a very diversified diet depending on location. Remnants of crustaceans, insect larvae, vegetation and fish have been found in the stomach of the white perch [15].

The intake of tritium through food may be an important element in the incorporation of tritium into the organically bound state. Evidence of this effect has been shown to exist in studies of cows' milk and tadpoles [16,17]. If fish were exposed to relatively high tritium concentrations, whether through water or food at some time in the past, the tritium incorporated in tissue will be lost by exchange or catabolism at a much slower rate than the turnover of body water [18]. This might also contribute to the large ratio of tritium in the bound to free state observed in this study.

Conclusion

In the aquatic environment, there is significant incorporation and retention of tritium in the bound state of bottom sediment and biota. Tritium expressed in terms of pCi/l was found to be, on the average, ten times higher in sediment, four times higher in fish and three times higher in rooted aquatic plants than the ambient tritium concentration in water. In some instances, the tritium content of the free or loose water of these components was also one to two times higher than ambient water.

The exchange of tritium for protium and the incorporation of tritium by metabolic processes in organic and mineral matrices will depend on the specific activity of the source of tritium, the form of tritium, and the magnitude and direction of the "isotope effect" in particular reactions. It will also depend on the amount of exchangeable hydrogen and growth stage of the organism. The greater mass of tritium compared to protium will affect chemical equilibrium and rate constants where molecules are labeled with tritium. The stability of the tritium-carbon bound in organic compounds will result in greater retention of tritium in these compounds [19].

In the environment, the exposure to tritium has been chronic and at levels that have been four to eight times higher than present concentrations. This permitted nonexchangeable sites to be labeled by tritium during formation, producing an "inheritance" of tritium that has persisted in sediment, detritius, fish and plants. This tritium, bound in an organic or mineral matrix, provides an additional source of tritium in the ecologic cycle. In this form, it appears to be an effective source of tritium to be synthesized in new organic material. Thus, rooted aquatic plants can acquire tritium from the sediment and fish can acquire tritium through the food web in addition to the tritium obtained from the water. In this way, it is possible that tritium can be accumulated in biota above current ambient concentrations.

ACKNOWLEDGMENTS

This investigation was supported by Contract AT (30-1) 4178 from the United States Atomic Energy Commission and is part of a New York University Medical Center Program supported by the Public Health Service, Division of Environmental Health Sciences, National Institutes of Health under Grant No. ES 00260.

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TRITIUM INTAKE IN NEW YORK CITY

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Tritium has been considered one of the least hazardous of the radioactive isotopes, partly because of its low energy beta-particles of 18 keV maximum and because the maximum penetration range is 5 μm for unit density matter. Also, the biological half-life is relatively short (8 to 14 days for man) and large doses are tolerated by experimental animals. Woodard [1] has stated that about two percent of the tritium intake, as tritiated water, is incorporated into tissue compounds and that the biological half-lives are much longer, ~20 to ~400 days. According to Woodard, incorporation of tritium as tritiated water into body components is rapid, and tritium is observed in two compartments. The first compartment is tissue free water tritium (TFWT) which is removed from the sample by mild distillation techniques or lyophilization. The second compartment is the dried tissue which is ashed and the water of combustion measured for tissue combined tritium (TCT). In our dietary study, we freeze-dried the samples to obtain the free water tritium and then the dried residue was combusted and the water of combustion measured for the combined tritium.

In view of the increase in the number of reactors and fuel reprocessing plants, it is now appropriate to establish base line amounts allowable for exposure to tritium by the general population [1,2]. Therefore, three intake categories, diet, air and fluids, were analyzed for their tritium concentration. The equipment utilized in this study included a freeze-drier (Table Size Atmo-Vac, Refrigeration for Science, Oceanside, NY); combustion apparatus (an induction furnace); electrolysis apparatus; dioxane scintillation solution composed of 7.8 g 2,5-diphenyloxazole (PPO), 0.2 g p-bis(o-Methyl-styryl)-Benzene (bis-MSB), and 100 g naphthalene dissolved and diluted to one liter with spectro-grade dioxane; and a Packard Tri-Carb Liquid Scintillation Counter, Model 3310.

Experimental

Diet

The food analyzed in this diet study was purchased during the winter and spring 1970-1971 at local stores. Only the edible portions were used for analysis. The samples were freeze-dried to remove and collect the loose water from the bulk sample. We evaluated the efficiency of this freeze-drying method for retaining all the condensible water by using known volumes of frozen water as a starting material. Recovery in all tests was greater than 99 percent. The loose water fraction was distilled to remove any traces of contamination and subsequently electrolytically enriched. The organic residue from freeze-drying is pelletized and ashed in an induction furnace. Approximately 50 g of dried sample are combusted at one time and, generally, it is necessary to combine three combustions to have a sufficient amount of water for electrolytic enrichment. Evaluation of the combustion system indicated that about 95 percent of the hydrogen in organic samples is converted to water. These tests were performed with oxalic acid and hydroquinone. Coincidentally, we were able to show that the combustion apparatus showed no memory effect at the activity levels of the samples we were processing.

Water

Tap water was distilled to remove impurities and electrolytically enriched prior to counting.

Air Samples

Water in the ambient air was condensed in a dry ice-acetone bath. The condensed water was distilled and electrolytically enriched prior to counting.

Enrichment

The procedure is an adaptation of that reported by Cosolito [3]. Electrolysis proceeds in an acid medium with platinum electrodes, and the cells are immersed in a cooling bath during operation. At the Health and Safety Laboratory (HASL) a platinum sheet is used instead of coiled wire, as the sheet is more resistant to deformation during handling. The cooling bath is maintained between 2° and 4°C and 0.7 ml H_2SO_4 are added to the 50-ml sample aliquot in the cell as electrolyte. The electrolysis is performed at 3 amp, resulting in a current density of 208 mA/cm^2. Under these conditions, the electrolysis proceeds at a rate of close to 1 ml/h. Aliquots of the sample are taken prior to and following electrolysis and titrated to determine volume reduction factors by changes in the acid concentration. Final volumes usually range from 5 to 8 ml. The enriched samples are vacuum distilled to remove the electrolyte and are subsequently measured bv liquid scintillation counting.

One or two standard solutions are usually processed with the samples. Since the electrolysis cells are connected in series, and the electrodes are of the same surface area, the enrichment factors obtained for the standards will also be applicable to the samples. The original tritium concentration in the sample is computed from the standard values and the volume reduction factors. Necessary calibration data are obtained from standard solutions and are periodically updated to incorporate the latest data. The "working curve" is an equation of the type $y = ax + b$ where y is related to the enrichment and x to the volume reduction. The calibration is reproducible to about five percent.

Counting

After electrolysis, liquid scintillation counting is performed on 1- or 2-ml aliquots of the enriched sample. Normally the samples are counted three times for 100 minutes and the count rates averaged. Counting standards and blanks are run concurrently with the samples. In addition, duplicates and standards are processed routinely in a quality control program. The background and efficiency are 8 cpm and 27 to 30 percent, respectively. The calculated detection limit is 0.9 dpm/ml assuming a one sigma background error, a 100-minute counting time, and a 2-ml sample volume. The detection limit for the enriched samples is about 0.1 dpm/ml.

Results and Discussion

The dietary tritium intake values determined for the nineteen food categories, for both loose water and combined tritium, are listed in Tables 1 and 2. The weight fractions of loose water and freeze-dried solids for each category were determined experimentally, and these results are in general agreement with Spector's [4]. The hydrogen content of the solids was determined by combustion. Using estimates of average yearly consumption [5], the total daily tritium intake presented in Table 3 was found to be 1700 pCi/day or 0.62 μCi/a. The Public Health Service has reported the dietary tritium intake [6] as 1200 pCi/day which is in general agreement with our results.

It is apparent that certain food categories exhibit some tritium enrichment in the organic fraction when compared to the loose water fraction. Table 4 lists the enrichment factors (ratio of the specific activity of the organic tritium to that of the loose water) for fourteen food categories. The average result for these values is 2.9. Evans and Cohen [7,8] have observed this enrichment phenomenon at environmental tritium concentration and have indicated that the enhancement is due to nonequilibrium conditions.

The tritium results for air samples are listed in Table 5. Some individual samples had to be composited because of the low water vapor content of the air in the winter. The overall average tritium concen-

Table 1

Dietary Tritium Intake

Category	Loose Water				Combined Water				
	Consumption (kg/a)	Water (kg/a)	Activity (pCi/l)	T Intake (µCi/a)	Solids (kg/a)	% Water	Water (kg/a)	Activity (pCi/l)	T Intake (µCi/a)
Carbohydrate Foods:									
Bakery Products	36.3	8.0	680	0.005	28.3	52	14.7	3050	0.045
Whole Grain Products	10.9	2.7	620	0.002	8.2	59	4.8	2400	0.012
Fresh Vegetables	43.2	37.2	1260	0.047	6.0	52	3.1	3110	0.010
Canned Vegetables	21.9	18.8	710	0.013	3.1	41	1.3	2490	0.003
Root Vegetables	16.9	14.2	580	0.008	2.7	50	1.4	2530	0.004
Potatoes	44.6	36.6	780	0.028	8.0	51	4.1	2850	0.012
Fresh Fruit	67.9	57.0	810	0.046	10.9	53	5.8	2430	0.014
Canned Fruit	15.0	12.9	710	0.009	2.1	53	1.1	1330	0.001
Juice	10.1	8.7	150	0.001	1.4	45	0.6	840	0.001
				0.16 µCi/a					0.10 µCi/a

Table 2

Dietary Tritium Intake

Category	Loose Water				Combined Water				
	Consumption (kg/a)	Water (kg/a)	Activity (pCi/l)	T Intake (µCi/a)	Solids (kg/a)	% Water	Water (kg/a)	Activity (pCi/l)	T Intake (µCi/a)
Protein Foods:									
Meat	72.3	41.9	680	0.028	30.4	78	23.7	970	0.023
Poultry	16.9	12.0	510	0.006	4.9	66	3.2	720	0.002
Eggs	16.0	12.6	190	0.002	3.4	65	2.2	840	0.002
Fish	8.1	6.6	150	0.001	1.5	56	0.8	840	0.001
Shell Fish	1.4	1.1	330	0.0003 0.04 µCi/a	0.3	52	0.1	750	0.0008 0.03 µCi/a
Dairy Foods:									
Milk	214	192.6	1040	0.200 0.20 µCi/a	21.4	65	13.9	1260	0.018 0.02 µCi/a
Dry Foods:									
Flour	43.1				43.1	52	22.4	2850	0.064
Macaroni	3.0				3.0	50	1.5	2920	0.004
Rice	2.6				2.6	53	1.4	810	0.001
Dry Beans	2.7				2.7	50	1.4	3270	0.005 0.07 µCi/a

Table 3

Dietary Tritium Intake bv Food Categories

Category	Tritium Intake (μCi/a)	
	Loose Water	Combined Water
Carbohydrates	0.16	0.10
Proteins	0.04	0.03
Dairy (Milk)	0.20	0.02
Dry Foods		0.07
Total:	0.62 μCi/a 1700 pCi/day	

Table 4

Ratio of Specific Activities of Loose Water and Organically Bound Hydrogen

Category	Enrichment
Fresh Fish	5.6
Juice	5.6
Bakery Products	4.5
Root Vegetables	4.4
Eggs	4.4
Whole Grain Products	3.9
Potatoes	3.7
Canned Vegetables	3.5
Fresh Fruit	3.0
Fresh Vegetables	2.5
Canned Fruit	1.9
Meat	1.4
Poultry	1.4
Milk	1.2
Average of Fourteen Food Categories	2.9

Table 5

Tritium Concentrations in Air Samples

Sampling Dates	nCi/l
April 16, 1970	0.82 ± 0.08
May 21, 1970	0.95 ± 0.08
June 8, 1970	0.77 ± 0.08
July 21, 1970	0.99 ± 0.08
August 27, 1970	0.93 ± 0.06
September 2, 1970	0.78 ± 0.06
September 14, 1970	0.59 ± 0.06
September 22, 1970	0.38 ± 0.06
October 1, 1970	1.25 ± 0.07
October 8, 1970	0.72 ± 0.07
October 15, 1970	0.63 ± 0.07
October 23, 1970	0.76 ± 0.07
November 2, 1970	0.54 ± 0.06
November 9, 1970	0.40 ± 0.05
November 25, 1970	1.75 ± 0.10
December 2, 1970	1.09 ± 0.06
December 14, 1970	1.49 ± 0.07
January 7, 10, 13, and 21, 1971	0.99 ± 0.07
January 28, 1971 and February 3, 11, and 19, 1971	1.46 ± 0.06
February 25, 1971	1.26 ± 0.07
Average:	0.93

Table 6

Tritium Concentration in Tap Water

Date	nCi/l	
May 1970	0.59	0.06
	0.41	0.06
	0.66	0.08
	0.41	0.07
June 1970	0.63	0.07
	0.58	0.07
	0.53	0.06
	0.49	0.06
July 1970	0.54	0.06
	0.46	0.06

tration is calculated to be 930 pCi/l. Assuming a 50 percent relative humidity and 55°F average temperature in New York City, the water content of the air would be 5.5 X 10^{-3} g/l. Assuming a total daily inhalation of 2 X 10^{4} liters, the intake of tritium from air would be 0.1 nCi/day or 37 nCi/a.

Tap water values are reported in Table 6. The average tritium concentration was 530 pCi; assuming a 1.70 liter daily water intake [9], the tritium intake would be 900 pCi/day or 0.33 µCi/a.

Therefore, the total tritium intake would be 2700 pCi/day or 0.99 µCi/a for the three modes of intake:

Diet	0.62 µCi/a
Air	0.037 µCi/a
Fluids	0.33 µCi/a

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CHAPTER NINE
Application of Tritium

TRITIUM IN BIOMEDICAL RESEARCH AND CLINICAL MEDICINE

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The first isotope systematically used for biological studies prior to 1940 was deuterium [1]. Tritium, the radioactive isotope of hydrogen, was employed in biology comparatively late. In 1951, Fitzgerald first used tritium for autoradiography [2]. Today, tritium is the most widely used tracer for biological research and is increasingly applied in medicine. The unique and special properties of tritium in biomedical research and clinical medicine are:

1. Ubiquitous nature of its isotope hydrogen in organic compounds.
2. The ease with which it may label any compound at particular molecular positions.
3. Its low energy beta-particle that is so useful for autoradiography.
4. Its decay to the chemically inert helium which converts its bonding partner carbon to a positively charged ion.

The use of tritium labeled compounds in biomedical research has been extensively discussed in 1967 [3]. A number of reviews and special articles on techniques have been supplied by manufacturers of equipment made especially for work with tritium labeled substances. The review presented here aims at focusing attention on some recent novel developments, including improved techniques and more refined experimental data, which also may potentially enhance medical diagnosis and therapy.

Specifically, the following subjects shall be discussed:

1. Specificity of labeling and metabolic hydrogen transfer reactions.
2. Labile hydrogen and macromolecular structure.
3. Tritium labeled proteins.
4. Tritium labeled DNA and cell proliferation.
5. Tritium labeled DNA for the purpose of cell killing.
6. Tritium, a tool for inactivating biologically relevant compounds.

Specificity of Labeling and Metabolic Hydrogen Transfer Reactions

In 1957, the Wilzbach technique for labeling compounds with tritium by exposure to tritium gas was notable for providing the researcher with a multitude of tritiated molecules [4]. The disadvantage of the technique is the lack of specificity of labeling and extensive molecular damage during the labeling process. These shortcomings do not apply to chemical labeling methods, nor to enzymatic and biosynthetic tritium transfer techniques. Not only do these allow the introduction of tritium to selected molecular sites, but they also may yield compounds with a high specific activity. Specificity of tritium labeling is a prerequisite for studying easily-traced reactions involving hydrogen transfer. On the other hand, if transfer reactions involving tritium are not properly recognized, experimental data are easily misinterpreted. It is thus important not only to test for radiochemical purity of the tritium labeled compound but also to reflect on biological stability; all possible metabolites of an organic compound should be considered and checked for labeling intensities.

An example is given by the widely used specifically tritiated nucleic acid precursors. Thus, tritium in the 5 position of uracil is lost when this pyrimidine ring is methylated to thymidine by thymidine synthetase. Moreover, tritium in this 5 position is also lost chemically by hot acid treatment often used for differential extraction of nucleic acids from tissues. Tritium in the 6 position of the uracil ring is stable under such conditions. Commercially supplied tritiated uridine does not always conform to the specifications of the manufacturer. So-called generally tritiated uridine was repeatedly found to contain less than five percent of the tritium in the stable 6 position and more than 95 percent in the labile 5 position [5]. It is obvious that 5-tritiated uridine is a more efficient precursor, especially for RNA labeling [7] and for DNA cytosine [8], whereas the 6-tritiated uridine is also a precursor for DNA thymidine.

Tritium may be transferred with metabolites such as monocarbon units [9]. Also, larger molecular groups may be involved and at times are not easily recognized. A recent report pointed out that diisopropylfluorophosphate (DFP) labeled with tritium was less efficient in tagging granulocytes of the peripheral blood than was the ^{32}P labeled compounds. The difference in labeling efficiencies may be explained by a loss of tritium carrying moity from the DFP after it was taken up by the cells. This argues against using tritium labeled DFP and recommends ^{32}P labeling instead [10].

The recovery of single specific metabolites of tritiated compounds is, of course, quite useful for quickly measuring reaction rates. For example, this is evident in the study of lipid metabolism, such as with unsaturated fatty acids specifically tritiated at the C atoms forming the unsaturated bonds [11]. It is simple to rapidly assay tritiated water generated by metabolism of specifically tritiated compounds. *In vivo* oxidation of 17-alpha-tritium-estradiol to estrone leads to loss of tritium from the steroid and to the generation of tritiated water. The tritium content of the body water, the urine, or respired air is then a measure of the rate of oxidation of estradiol [12]. This technique is of considerable interest to clinical medicine and is likely to be applied to other *in vivo* reactions involving loss of protons.

A great number of molecules have been labeled *in vitro* by hydrogen transfer reactions catalyzed by pure or by crude enzyme preparations. Complex molecules such as cholesterol [13] and less complex compounds such as glutamate [14] have thus been specifically tritiated. *In vivo*, similar reactions with tritium permit the differentiation of different hydrogen pathways. For example, rat liver mitochondria were allowed to catalyze oxidative phosphorylation in the presence of tritiated water and oxygen-18. When the mitochondria were rapidly chilled after the reaction, they were shown to retain small quantities of tritium dependent upon the temperature of incubation.

Tritium binding was different when the tritium donor was not water but 2-tritium-glutamate. Nonspecific hydrogen exchange in mitochondrial protein probably occurred independent of the proton binding from oxidative phosphorylation demonstrated by the tritium transfer from 2-tritiated-glutamate [15]. Hydrogen pathways are especially manifold from the various positions of carbohydrates [16,17]. Thus, glucose-6-tritium together with glucose-6-^{14}C as internal standard was used to study the synthesis of RNA, DNA and protein in tumor slices [18]. This method is promising to quickly and simultaneously test cytotoxic drugs for various metabolic reactions.

Tritium transfer reactions are influenced by isotopic effects which may be particularly large since tritium is about three times as heavy as normal hydrogen. These isotopic effects are indeed useful for determining rate limiting steps in complex metabolic reactions involving multiple enzymes, and for differentiating various molecular sites involved in a reaction [19-21]. For example, it was shown that the coenzyme B$_{12}$ donates hydrogen to the reaction product from at least three different positions and may form with the enzyme dioldehydrase an undissociable complex [22].

There is always exchange of tritium in body water with hydrogen in tissue solids [23]. Yet no net effects from isotopic discrimination were recognized. A long-term study with kangaroo rats living in a tritium contaminated environment indicated that the specific activity of tritium in tissue solids was approximately the same as that of tritium in body water [24].

Tritiated water is widely used in clinical medicine for measuring total body water and water turnover. More recent improvements include the simultaneous measurement of other body compartments by means of multiple isotope assays [25-28].

Labile Hydrogen and Macromolecular Structure

Hydrogens forming intermolecular bonds, such as in proteins and nucleic acids, may exchange with tritium. This hydrogen-tritium exchange offers a most valuable technique for studying macromolecular structure and the effect of structural changes on specific functions. The analysis of tritium-hydrogen exchange on Sephadex has become especially useful [29]. The rate of hydrogen-tritium exchange measured on a Sephadex column permitted the recognition of subtle and transient changes in the secondary structure of transfer RNA [30]. Also, DNA possesses rapidly and slowly exchanging hydrogens involved in base pairing [31]. Tritium exchange measurements were interpreted to indicate that a single base, situated between fixed upper and lower neighboring bases, may rotate around its phosphodiester bond [32]. During the process of transcription, hydrogen exchange increases, which supports the hypothesis that the DNA template is temporarily denatured by the action of RNA polymerase [33]. Also interaction of macromolecules with certain trace metals causes conformational changes. Such studies were made with the help of the tritium-hydrogen exchange technique with conalbumin (ovo-transferrin) [34], collagen [35] and insulin [36].

Tritium-Labeling Proteins

Important for endocrinological and immunological studies is the availability of tritiated proteins which are biologically competent and

and which also have a high specific activity for autoradiographic analyses. The replacement of hydrogen by tritium in a protein molecule does not necessarily lead to a loss of molecular function. This is especially pertinent to investigating immune reactions. Various radioimmune assays with tritiated compounds such as plasma estrogens [37] or insulin [38] are useful in clinical medicine.

A number of techniques for tritium labeling of antigens, proteins and polypeptide hormones were reviewed in 1969 [39]. Labeled protein is readily obtained by biosynthesis using tritiated amino acids. A recent study succeeded in labeling *Trichinella spiralis* larvae antigen with tritium by 13 labeled amino acids as precursors. The tritiated *Trichinella spiralis* larvae antigen had full biological specificity and was capable of transforming sensitized lymphocytes [40]. Hydrogen exchange *in vitro* was used to tritiate thyrotropin that kept its biological activity. Only the terminal galactose residue of the molecule was tritiated in this work, first by exposure of the hormone to galatose oxidase and then by reduction with tritium-sodium borohydrite [41]. Oxytocin, one of the pituitary hormones, was tritiated to a specific activity of 36 Ci/mM by a novel two-step method. The tyrosine ring was selectively iodinated under mild conditions leaving disulfide bridges unaffected. The thus iodinated hormone was biologically inactive. When the hormone was then allowed to react catalytically with tritium, the peptide-bound iodine was exchanged for tritium and the biological hormonal activity recovered fully [42]. Other chemical labeling procedures such as acetylation and alanylation have become useful. Mouse alloantibody (gamma globulin) was tritiated by the reaction with tritium labeled DL-alanine-N-carboxylanhydride [43], and tritiated acetic anhydride was used to prepare tritiated gamma globulins [44] or tritiated trypsin with a specific activity of 1.4 Ci/mM [45].

Tritium labeling of proteins with preservation of biological activity may also be achieved with the free radical interceptor method alone or combined with other methods. Accordingly, lysozyme in the dry state was gamma irradiated *in vacuo* to produce carbon free-radicals which reacted with tritiated sulfide present during the irradiation. The tritiated lysozyme was chromatographically indistinguishable from the native enzyme. It is noteworthy that the site of the free-radicals produced in the protein can be analyzed from the tritium distribtuion between the various amino acids. This allows the observation of secondary free-radical distribution in the irradiated dry protein [46]. Secondary free-radical distribution is also an important phenomenon for the interpretation of consequences of tritium transmutation to helium, which inevitably leads to the production of a positively charged radical carbonium ion at those sites where the tritium was bound. The tritium gas exposure technique for labeling proteins and peptides was further developed. Modified techniques led to useful tracer [47,48] in the case of mucopolysaccharides [49].

Tritium Labeled DNA and Cell Proliferation

For studying DNA synthesis and metabolism in intact cells and for investigating cell proliferation, there is no other tritiated precursor more eminent than tritiated thymidine. DNA is easily labeled by biosynthesis. Chemical tritiation by exchange reactions [50-52] was also used and yielded tritiated DNA that retained properties comparable to native DNA. Such techniques are perhaps useful when labeling of DNA by biological synthesis is inconvenient. The tritium gas exposure technique is not optimal for tritiating DNA because it induces excessive molecular decomposition [53].

Tritiated thymidine was introduced in 1957 by Taylor, Woods and Hughes from Brookhaven [54] and independently by Verly and Hunebelle in Belgium [55]. The particular advantage of tritiated thymidine in comparison with ^{14}C labeled thymidine is the high specific activity and the low energy beta that can be registered autoradiographically and which can localize tritium within subcellular structures. The autoradiographic resolution for tritium is 0.6 μ in the light microscope [56] and about 0.1 μ in the electron microscope [57]. A vast number of studies has appeared since the early work at Brookhaven [58,59]. The kinetics of cellular proliferation have been worked out in many cellular systems throughout nature, and many discoveries on the mechanism of synthesis and metabolism of DNA at the cellular level were made. This work has been repeatedly reviewed [3,60,61].

With respect to studies on cell proliferation, a novel method pertains to autoradiographically recognizing the contribution of cells with a long generation time to the steady state of cell populations with short generation times. All cells of rat embryos were labeled *in utero* by continuous infusion of tritiated thymidine into the pregnant animals. In the newborn rats, the rapidly renewing cell systems are depleted of the label within days, and those labeled cells which have a long life span are left behind. In an autoradiogram, these cells are then easily classified, and their reactions, for example, towards injury can be readily observed [62,63]. These studies gave strong evidence for the function of the bone marrow lymphocytes as stem cells.

An important problem in work with tritiated thymidine is posed by reutilization of thymidine from dead cells for DNA synthesis by living cells. Long-term observations of thymidine labeled cells may thus be misleading. Thymidine reutilization can be corrected for by comparing the turnover rates of cells tagged with tritiated thymidine with those labeled with iododeoxyuridine. This precursor is an analogue to thymidine but is inefficiently utilized and also inefficiently reutilized [64-67].

A useful double labeling method applies to directly determining the length of the DNA synthesis time of cells and the rate of cell proliferation. Here thymidine is labeled either with tritium or with ^{14}C. The two differently labeled thymidines are sequentially administered to the cells either *in vivo* or *in vitro*, and the relative numbers of tritium and ^{14}C labeled cells in the specimen are autoradiographically determined and used for computation of kinetic parameters [68-71]. This technique appears suitable in clinical medicine for quickly examining cell kinetics in tissues obtained by biopsy [72,73]. In combination with tritiated thymidine, other labeled substances may be used, for example, specifically tritiated carcinogenic azo dyes [74] or croton oil [75] with high specific activities for studying drug mediated induction of tumors.

Cells labeled for their life span with tritiated thymidine can greatly aid transplantation studies. Thus, the proliferation of transfused labeled bone marrow cells may be autoradiographically observed to differentiate in nonlabeled hosts [76,77]. Single labeled cells may also be seen to differentiate in millipore chambers incubated under proper conditions [78], or within clones developing from single cells [79]. Similarly, embryonic tissue labeled with tritiated thymidine may be grafted into other embryos and watched for differentiation, as it was done, for example, recently with chick endomesoderm transplanted into the cardiac area of another embryo in the same stage. The transplanted cells labeled with tritiated thymidine were autoradiographically recognized to produce cardiac tissue [80].

Also, the fate of single tritium labeled chromosomes taken up by mammalian cells could be observed [81]. The entrance of single chromosomes into mammalian cells might lead to integration of undegraded DNA nucleotide chains into the host genome, as it is known to occur in microorganisms, where the transforming principle of DNA has been firmly established. Thus not only viral DNA but also homologous DNA nucleotides, or, in other words, genetic information, may under particular conditions be exchanged between cells. Strong evidence for this to occur physiologically in lymph nodes has recently been obtained [82].

Tritium-Labeled DNA for the Purpose of Cell Killing

The low energy beta-particle of tritium incorporated into the cell nucleus, for example with tritiated thymidine, irradiates mainly this nucleus, and the effects produced are comparable to the effects from external X-irradiation. A cell nucleus of 8 μ diameter uniformly labeled with tritium, for example in DNA, will absorb an average of 0.3 rad per tritium decay in the nucleus [83]. After tritiated thymidine was first synthesized at Brookhaven, it was tested for killing selectively DNA labeled cells [84]. By now the technique of cell suicide, induced by tritiated thymidine incorporated into the cellular DNA, is a most useful tool in research of stem cell kinetics [85-87] or the production of immunologically competent cells [88,89]. It was indicated by this technique that antibody producing cells are derived from a cell population that has at least a temporarily rapid turnover. A high dose of tritiated thymidine was lethal to antibody forming cells in spleen cell cultures only 24 to 32 hours after an antigenic stimulus.

The suicide technique was also instrumental in determining, for the first time, *in vivo* variations in radiosensitivity during the generation cycle of stem cells in mouse bone marrow [90] and in fetal liver [91]. When, for example, resting colony-forming units in mouse bone marrow were stimulated into proliferation, the overall radiosensitivity of that cell population was not altered. However, when the cells in DNA synthesis were eliminated by tritiated thymidine suicide, the remaining cells were significantly less radiosensitive. This indicated that *in vivo* "S-phase" cells capable of forming spleen colonies were more radiosensitive than such cells in other stages of the cell cycle, including G_0.

Differential radiosensitivity of various cellular systems, may be studied in rodents, for example, continuous infusion of tritiated thymidine into pregnant females. This leads to widespread labeling of all cells in the fetal tissues, some of which cease to proliferate after birth and during adolescence [92,93]. Depending upon the time of commencement of the continuous infusion of tritiated thymidine, particular organs developing in the embryo are thus amenable for selective studies.

Interesting, in this context, is the selective binding within tumor cells of vitamin K derivatives heavily tritiated in order to attempt a specific tumor therapy [94,95]. Other tumor binding agents labeled with tritium are discussed in other reports in this book.

Tritium as a Tool for Inactivating Biologically Relevant Compounds

The toxicity of tritium largely depends on its position within the cell and may involve a transmutation effect. This was discussed in more detail in the context of the tritium transmutation problem [96]. It was emphasized that tritiated RNA and protein precursors cause less lethality than tritiated thymidine, which is incorporated specifically into DNA. However, this could be ascribed to the geometry of the beta-particle originating in these various compounds. With DNA being held as the radiosensitive site within a cell, it is noteworthy that mitochondrial DNA synthesis appeared to be less radiosensitive than nuclear DNA synthesis [97].

The effect of tritium transmutation on DNA could be documented. Transforming DNA from pneumococcus was labeled with tritiated thymidine only in one of the two complementary strands [98]. Tritium decays in DNA preparations, which were kept in the frozen state, produced four to five times more breaks in the labeled strand than in the unlabeled one. By thus selectively inactivating single DNA strands, it was possible to prove that only single DNA strands are indeed integrated into the host pneumococcal DNA, and that one single strand break inactivates the transforming activity of the donor DNA. Thirty percent of the tritium decays produced single strand breaks, and less than one percent produced double strand breaks.

On the other hand, a specific mutation of lactose operator gene in E. Coli was produced by incorporating various tritiated amino acids into the regulator protein, which is locus specific for the DNA constituting the lactose operon [99]. The effect is not as yet fully explained. It may be due to irradiation, and the transmutation effect could also be involved in leading to a diffusion of radicals from the site of isotopic decay to directly neighboring molecules bound by weak interactions.

Conclusion

This short review of the particular applications of tritium in biomedical research and clinical medicine has emphasized that this tracer has achieved an irreplaceable position in modern biomedical technology. It was only 20 years ago that tritium first came into use. Now it is a star in its own right, with virtues widely applauded and with faults that ask for more definition and final avoidance. To elucidate the variety of problems associated with tritium, the tritium symposium was convened. It has been this author's contribution to emphasize more virtues than faults that characterize tritium, which is analyzed in more detail in the accompanying articles.

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TRITIUM TRACERS IN BIOLOGY - FACTS OR FICTION?

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The remarkable versatility of tritium and the relative ease of labeling both simple and complex organic molecules with this radionuclide have resulted in the widespread use of tritium compounds as tracers in biological research. However, there are a number of properties peculiar to tritium compounds in tracer applications which should be recognized so "true" facts can be obtained even though a speculative interpretation may be placed upon them. Charles Darwin once said "false facts are highly injurious to the progress of science for they endure long life but false views if supported by some evidence do little harm for everyone takes a salutory pleasure in proving their falseness."

Some physical properties of tritium are summarized in Table 1. Although the difficulties in the measurement and detection of the weak beta radiation retarded an earlier development of tritium compounds as tracers, these problems have now been largely overcome. The weak beta energy coupled with the very high specific activities attainable with tritium compounds have made tritium tracers of great value in high resolution autoradiography. The low range (1 μm) of the tritium beta-particle in photographic emulsion permits the precise localization of the compound in a biological specimen. The high molar specific activity of tritium compounds also permits the use of almost physiological (chemical) concentrations of labeled drugs, for example, for *in vivo* tracer experiments.

Table 1

Some Physical Properties of Tritium

<table>
<tr><th colspan="2">Mass 3
Radioactive</th></tr>
<tr><td>Radiation</td><td>β^- (100%)</td></tr>
<tr><td>Half-Life</td><td>12.26 Years</td></tr>
<tr><td>Energy</td><td>18.6 keV (Max)
5.7 keV (Mean)</td></tr>
<tr><td>Specific Activity</td><td>29.12 Ci/μg atom of hydrogen (Max)</td></tr>
<tr><td>Range of Particles</td><td>4.5 to 6 mm in Air
6μm in Water
1μ in Photographic Emulsion</td></tr>
</table>

Tritium and its compounds are currently used both as tracers for hydrogen and, more frequently, as tracers for carbon structures. Consequently, very careful consideration must be given to the integrity of the carbon-tritium bond.

Two interrelated factors need to be clearly recognized in the application of tritium tracers to biological research. These are:

1) The importance of knowing the specificity of labeling in the tracer compound, i.e., the exact distribution of the tritium atom(s) in the labeled molecule, and

2) the stability of the tritium label under the experimental conditions.

The specificity of tritium labeling is normally related to the method used for preparing the tritiated compound. The stability of the tritium atom(s) in a labeled molecule is related both to the position of the label and to the experimental conditions under which the compound is being used.

Specificity of Tritium Labeling

Three general methods are in current use for the preparation of tritium compounds [1]:

1) Isotope Exchange Reactions - with tritium gas (Wilzbach method; radiation catalyzed), or with tritiated solvents (homogeneous

acid or base catalyzed; heterogeneous hydrogen transfer metal, Pt or Pd, catalyzed).

2) Chemical Synthesis - which include catalyzed hydrogenation or halogen-tritium replacement reactions, and the use of tritiated metal hydrides.

3) Biochemical Methods - involve the use of enzymes for the conversion of substrates labeled either by exchange or chemical synthesis into more complex labeled compounds.

Most isotope exchange reactions suffer from the disadvantage that "generally" labeled compounds are produced in which the position and distribution of the tritium atom(s) is(are) unknown. Chemical synthetic methods are therefore to be preferred. However, even some chemical methods which would be expected to yield specifically labeled compounds do not always result in products of clearly defined patterns of labeling [1,2]. Some examples will illustrate these points and will emphasize the need to know where the tritium atom(s) is(are) located in the labeled tracer in order to interpret the results with confidence.

1) Cytology - Tritiated thymidine (labeled in the 5-methyl or 6-position) as a tracer for DNA is currently in widespread use. The second most widely used radioactive tracer compound is uridine-5-T. The use of this labeled compound as a specific tracer for RNA depends upon its freedom from uridine-6-T. If the latter compound is present then some DNA labeling will result through the thymidine synthetase pathway even though the alternative minor pathway through 2'-deoxycytidine is blocked (Figure 1) [3,4].

Provided the catalytic halogen-tritium replacement reaction used to prepare the uridine-5-T is carried out quickly (10 minutes) more than 98% of the tritium is located at the 5-position. Prolonged reaction times (hours) can result in up to 15% of the tritium being present in the 6-position [4].

Figure 1

Uridine-5-T as a specific label for RNA

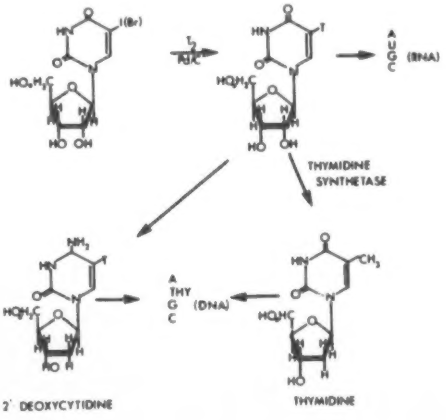


Figure 2

Metabolism of Vitamin-B_6-H3

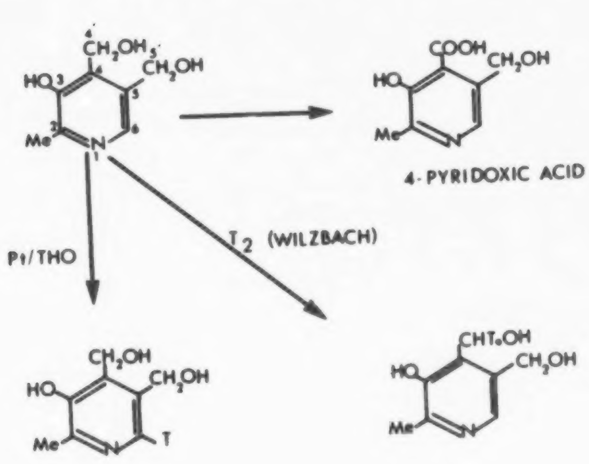


Figure 3

Preparation of tritiated folic acid

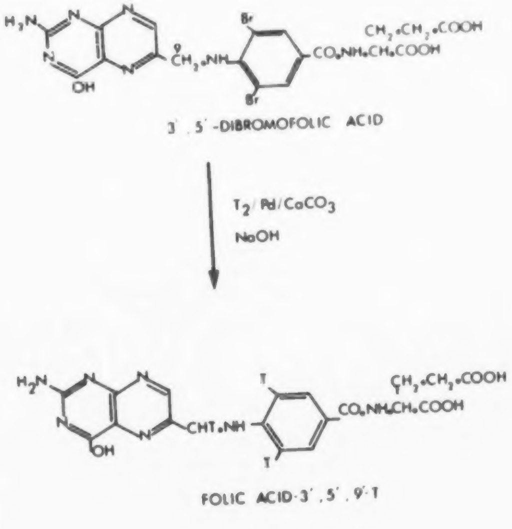


Figure 4

Biosynthesis of estrogens

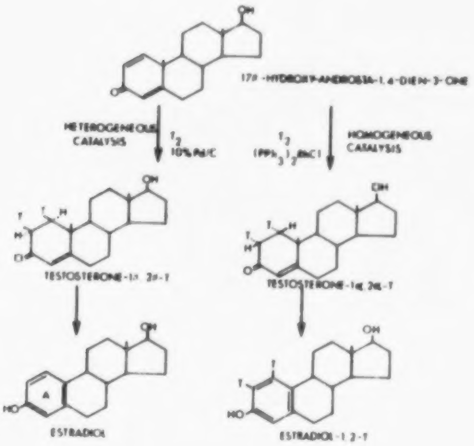
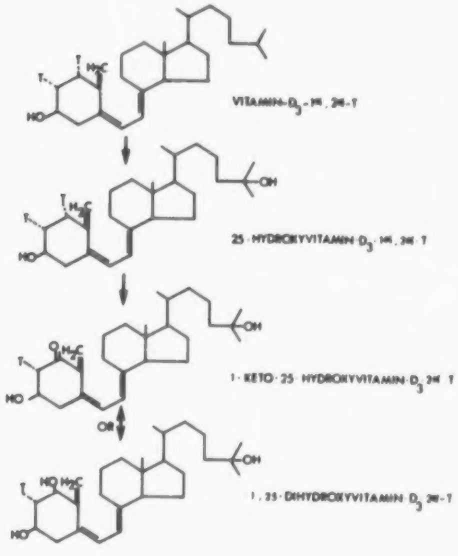


Figure 5

Metabolism of Vitamin-D_3 (Cholecalciferol) in man


T
T
H_2C
HO
VITAMIN-D_3-1α,2α-T
OH
25-HYDROXYVITAMIN-D_3-1α,2α-T
O
1-KETO-25-HYDROXYVITAMIN-D_3-2α-T
OR
HO
1,25-DIHYDROXYVITAMIN-D_3-2α-T

2) Metabolism Studies - Use of vitamin-B_6-T (pyridoxine) labeled by the Wilzbach method [5] results in loss of at least 60% of the label during metabolism to 4-pyridoxic acid, the major metabolite in man (Figure 2). Using pyridoxine-T labeled by catalyzed exchange in tritiated water, less than 10% of the tritium is lost under the same conditions, as most of the label is situated at the 6-position using this method [6].

For many years tritiated folic acid, believed to be labeled predominantly in the 3', 5'-positions, has been used for folic acid malabsorption studies and in the study of megaloblastic anemia [7,8]. In such studies, the fate of the whole molecule is followed and it does not matter where the tritium atoms are located provided they are stable under the experimental conditions. Recent investigations of folic acid metabolism indicated that only about 40% of the tritium is located in the 3', 5'-positions although the compound was prepared from 3', 5'-dibromfolic acid by a rapid catalytic halogen-tritium replacement reaction. Oxidation of the folic acid-T revealed 60% of the tritium located at the 9-methylene hydrogen positions (Figure 3).

Stereospecificity of labeling becomes important in metabolism and biological hydrogen transfer reactions. It has been found, for example, that during the conversion of androstenes to estrogens the 1β, 2β-hydrogen (tritium) atoms are eliminated in the aromatization of the steroid A-ring [10,12]. In studies of metabolism it is therefore important that the tritium atoms are in the 1α, 2α-configuration of the substrate androstenes so that the label is retained in the metabolites (Figure 4) [13]. Reduction of 3-oxo-$\Delta^{1,4}$-steroids with tritium and a homogeneous catalyst yields steroids labeled mostly in the 1α, 2α-positions.

Sometimes the loss or retention of a tritium atom can help in the identification of a metabolite. A recent example concerns the metabolism of vitamin-D_3. The tritium atom at the 1α-position of vitamin-D_3-T is lost during metabolism and about 50% of tritium is lost from vitamin-D_3-1,2-T [14, 15]. Since the tritium atoms are retained in the formation of the first metabolite 25-hydroxyvitamin-D_3, the tritium deficient metabolite was predicted to have a 1-keto- or 1-hydroxyl group. The metabolite has now been identified [16] as 1,25-dihydroxyvitamin-D_3 (Figure 5).

The use of stereospecifically labeled tritiated mevalonic acids for the study of hydrogen elimination reactions in the biosynthesis of steroids and terpenes offer other examples of the importance of stereospecific labeling with tritium [17].

3) Enzyme Assays - The presence or absence of certain enzymes in body fluids can sometimes be related to a pathological condition. The great sensitivity offered by the radiotracer technique has resulted in the continuing development of radiochemical methods for the assay of enzymes.

For example, tyrosine hydroxylase, an important enzyme in catecholamine biosynthesis, is determined by tritium release (as tritiated water) during the hydroxylation of tyrosine-3, 5-T (Figure 6) [18]. For accurate results it is, of course, necessary to know the specificity of the labeling.

An attempt to use a similar tritium release method for the assay of phenylalanine hydroxylase resulted in the discovery of an intramolecular hydrogen (tritium) transfer reaction, as the tritium atom in the paraposition of phenylalanine was retained in the

Figure 6

Assay of tyrosine hydroxylase

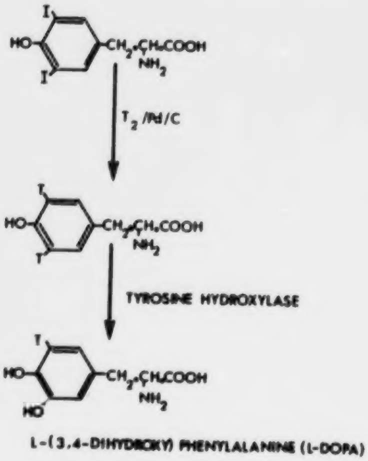


Figure 7

Assay of phenylalanine hydroxylase

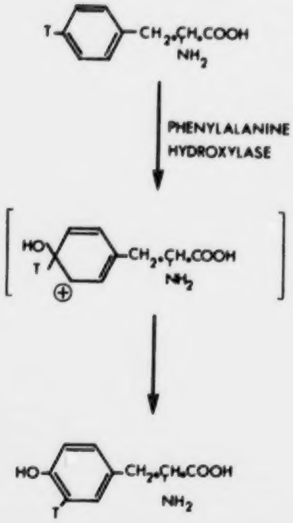
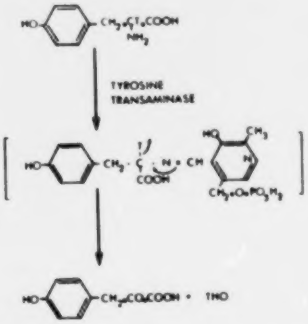


Figure 8

Assay of tyrosine transaminase



tyrosine formed (Figure 7). This is now the well-known "NIH shift" discovered at the National Institutes of Health, Bethesda [19], and is regarded as a fairly general phenomenon associated with the reaction of aromatic rings with hydroxylases.

The loss of tritium from the 2-position, i.e., label at the asymmetric center, of amino acids is used as a method for the assay of transaminases; for example, the use of tyrosine-2-T for the measurement of tyrosine transaminase (Figure 8). It is interesting to note that the assay of enzymes within a cell by tritium release (as tritiated water) into the medium has the unique advantage that cells do not have to be destroyed for measurement of the enzyme activity.

Determination of the Specificity of Labeling

Determination is not always easy to perform as noted by Simon and Floss [21], and is particularly difficult with tritium compounds where degradation methods are subject to error due to exchange reactions. Substitution reactions are preferred where applicable but even then one must be sure that no loss of tritium occurs by exchange reactions with the solvents. For example, the specificity of labeling in uridine-5-T is routinely checked by bromination. The 5-bromouridine isolated should be unlabeled if all the activity is in the 5-position (Figure 9) [4].

Routine measurement of triton magnetic resonance spectra (TMR) has been described recently for the first time [22] using a micro-bulb assembly of the type described by Flath *et al.* [23] to obviate radiological hazards (Figure 10). The proportionality between the integrated TMR signal intensity and the isotopic abundance has been demonstrated (Figure 11). An example of a typical TMR spectrum is shown for uridine-5-T (Figure 12); self-radiolysis products and uridine hydrate are easily observed. With the aid of a computer measurement on samples of tritiated compounds having an isotopic abundance of tritium at one position of 2% can readily be obtained. This *nondestructive* method for the determination of labeling patterns in tritiated molecules is considered a major advance towards solving this problem.

Figure 9

Specificity of labeling in uridine-5-H3

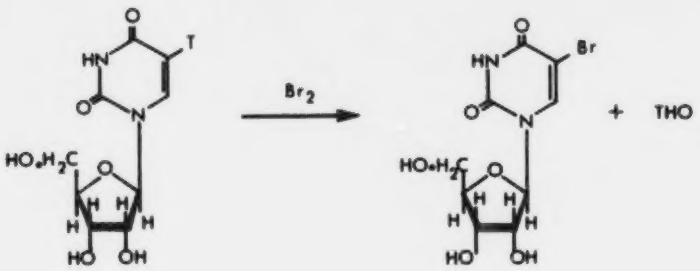


Figure 10

Micro-bulb assembly for TMR spectra. Micro-cell assembly showing: (a) micro-bulb, 'Teflon' holder and threaded insertion-tool; (b) micro-bulb, surrounded by CCl_4, in NMR tube.

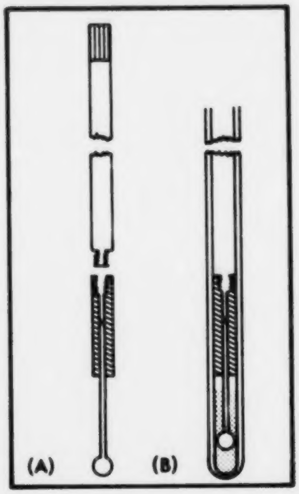


Figure 11

Proportionality between TMR spectra and isotopic abundance

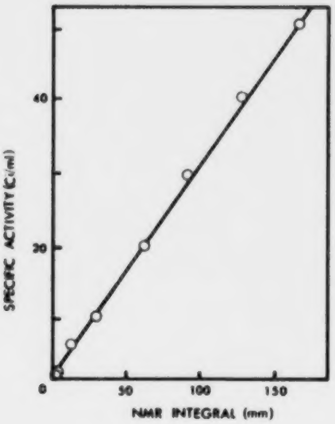
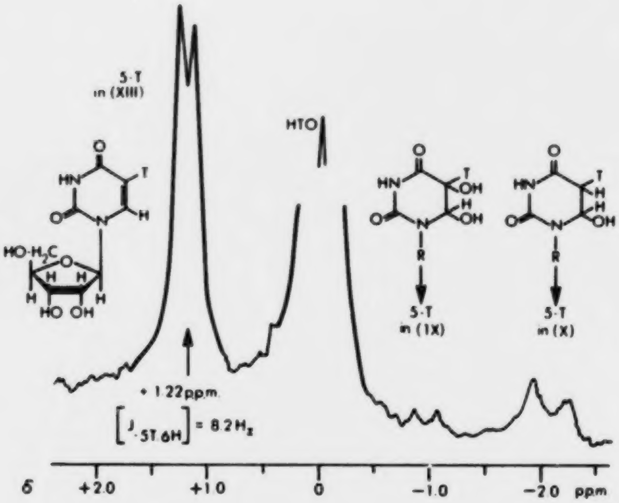


Figure 12

N.m.r. spectrum of uridine-5-T and decomposition products. 31Ci/mmol 0·16M Soln. in THO at 10Ci/ml. After 6 days (dose 18 Mr).



Stability of Tritium Atoms in Molecules

Many research investigators are reluctant to use tritium labeled compounds as tracers because of possible instability of tritium atoms in the labeled compound under experimental conditions. Four kinds of tritium labels can be distinguished:

1) Tritium label which is readily (often instantaneously) exchanged in hydroxylic solvents under neutral conditions. This is termed "labile" tritium and usually refers to tritium attached to N, O or S atoms.

2) Tritium label which is not exchanged under all conditions of pH and temperature, which does not result in decomposition of the molecule, and which requires the presence of a hydrogen transfer catalyst (chemical or biochemical) for exchange to occur in hydroxylic solvents. This is termed "nonlabile" tritium.

3) Less clearly defined tracer in which the tritium atoms are normally regarded as "nonlabile" but which can be exchanged with hydrogen atoms of a hydroxylic solvent under certain conditions of

Table 2

Tritium Exchange from Compounds in Dilute Aqueous Solution at +2°C [24]

Compounds	Specific Activity Ci/mmol	Storage Time weeks	Labile Tritium percent
L-Alanine-T(G)	0.17	75	<1
5-Aminolevulinic Acid-3,5-T	2.5	30	20
L-DOPA-2,5,6-T	34.7	11	15
L-Histidine-2,5-T	7.5	14	9
L-Leucine-4,5-T	1.0	25	1
L-Phenylalanine-4-T	1.0	21	2
L-Tyrosine (side-chain-2,3-T)	3.6	30	<1
L-Tyrosine-3,5-T	28.2	30	8*
Adenine-2,8-T	3.6	30	5
Adenosine-8-T(n)	3.1	12	6
Guanosine-8-T	4.9	16	13
Thymidine-(methyl-T or 6-T)	5.0	12	<2
Uridine-5-T	5.0	12	3

*some decomposition

pH, and temperature. This tritium is referred to as "acid-labile" or "alkali-labile" tritium. The rate of exchange of this "pH-labile" tritium will, of course, depend upon the actual pH and temperature of the solution. The rate of exchange will normally increase with temperature.

4) Tritium atoms which become labile under biological conditions may be termed "biolabile;" such tritium atoms may be quite stable under chemical conditions.

Prolonged storage of some tritium compounds even in neutral aqueous solution results in the loss of tritium from the compounds by isotope exchange [24]. Some examples are given in Table 2. Tritium atoms in aliphatic compounds are normally firmly bound under all conditions of pH unless present in enolizable positions, e.g., 5-aminolevulinic acid-3,5-T (Table 2). Tritium atoms in aromatic rings are also stable unless the ring has nucleophilic substituents. Compare, for example, L-phenylalanine-4-T and L-DOPA-2,5,6-T (Table 2). However, most of the difficulties in the uses of tritium compounds as tracers arise with compounds in the classes (3) and (4) where the tritium atoms are "acid-labile," "alkali-labile" or "biolabile." Some examples will illustrate these difficulties.

(1) Amino Acids. Transaminases, as impurities in crude D- (or L-) amino acid oxidases, caused unexpected loss of tritium from the alpha-position (asymmetric center) of generally tritiated DL-amino acids during oxidase reactions [25]. Some typical results are shown in Table 3.

Table 3

Effect of Oxidases on Tritiated Amino-Acids

Amino-Acid	Specific Activity mCi/mM	L-Amino-Acid Activity mCi/mM	D-Amino-Acid Activity mCi/mM	Tritium Loss %
DL-Alanine-T(G)	1540	20.5	-	99
DL-Alanine-T(G)	62	63	-	Nil*
Glycine-2-T	92.3	48	-	48
DL-Leucine-T(G)	170	95.6	-	44
DL-Methionine-T(G)	65.6	32.8	-	50
DL-Phenylalanine-T(G)	825	561	-	32
DL-Phenylalanine-T(G)	825	-	561	32
DL-Tyrosine-T(G)	101.4	61.5	-	39

*with purified D-amino-acid oxidase (Boehringer)

The observations with tritiated glycine led to the speculation of stereospecifically labeled glycines [1] which has recently been confirmed [26,27]. The loss of tritium from the asymmetric center is used with advantage for the assay of transaminases [25].

Note that instability of the label under certain chemical (pH and temperature) conditions does not necessarily preclude that tritiated compound from being useful for metabolism studies provided that careful control experiments are carried out. An example is L-3, 4-dihydroxyphenylalanine-(ring-2,5,6-T) (Figure 13) in which the tritium atoms are unstable to acid or alkaline [28], although the compound has been successfully used for metabolism studies in humans [29].

(2) Carbohydrates. The loss of tritium from the 2-position of glucose-T is reported as providing a valid method for estimating glucose utilization and production in vivo, since the labilization is irreversible [30]. Biological hydrogen transfer reactions

Figure 13

L-3,4-dihydroxyphenylalanine-(ring-2,5,6-H3).

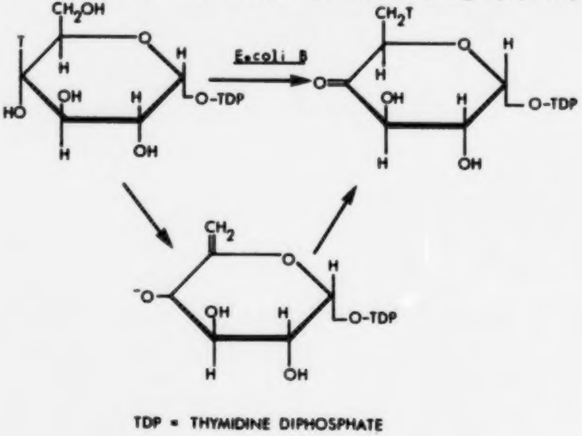
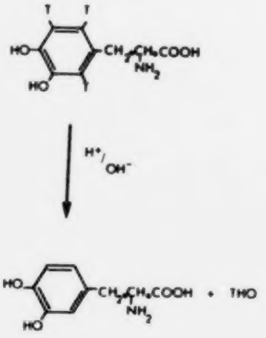


Figure 14

Formation of TDP-4-KETO-6-deoxyglucose-6-H3



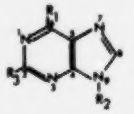
involving the NAD and NADP coenzymes normally proceed without exchange of the transferred atom with the aqueous medium. Thus thymidine diphospho(glucose-4-T) is coverted into thymidine diphospho (4-keto-6-deoxyglucose-6-T) without loss of the tritium atom (Figure 14) [31]. With thymidine diphospho(glucose-5-T) most of the tritium is lost by exchange with the aqueous medium, suggesting that a methylene intermediate is involved in the reaction [32]. Another example of the retention of the tritium label is the transfer of tritium from glycerol-2-T into fatty acids [33].

(3) "Nucleics." The stability of tritium atoms in the important classes of labeled compounds, purines, pyrimidines, nucleosides and nucleotides has been recently reviewed [34]. An important outcome of this work is the recognition of the relative ease of exchange of the tritium atom from the 8-position of the purine ring, under various conditions of pH and temperature, as compared to the 2-position.

Some kinetic data are given in the Table 4 and show that even in neutral aqueous solution the tritium atom in the 2-position of adenine undergoes exchange nearly 2000 times as slowly as that in the 8-position, which in turn exchanges at the same rate as the 8-position of purine [35]. Tritium in the 8-position of adenosine exchanges twice as fast as in adenine. Current investigations indicate that the tritium atom in the 8-position of guanosine exchanges faster than either adenine or adenosine (cf. Table 2) probably due to the influence of the 2-hydroxyl group.

Table 4

Exchange from tritiated purines in aqueous solution



COMPOUND	POSITION OF LABEL	pH	TEMPERATURE °C	TIME Hrs	RATE OF EXCHANGE k_t (s^{-1})	% LOSS OF TRITIUM
PURINE ($R_1 = R_2 = R_3 = H$)	8	7	85	•	3.2×10^{-5}	•
ADENINE ($R_2 = R_3 = H; R_1 = NH_2$)	2	7	85	•	5×10^{-8}	•
ADENINE	8	7	85	•	3.2×10^{-5}	•
ADENOSINE ($R_3 = H; R_1 = NH_2$; R_2 = ribose)	8(85%)	7	85	•	6.3×10^{-5}	•
ADENINE	2	7.4	100	1	•	6
ADENINE	8	7.4	100	1	•	48
ADENINE	2	8	37	24	•	3
ADENINE	8	8	37	24	•	3

Determination of the Instability of the Tritium Label

Chemical instability, for example, under various conditions of pH and temperature, can usually be determined by simple experimentation.

Biological instability cannot readily be predicted but can be determined in two principal ways:

1. By conducting a total activity balance and testing for the presence of tritiated water formed by exchange. If no "bio-labile" tritium is present in the substrate then it will be recovered at the same molar specific activity as before administration [6]. Likewise, metabolic products will have the same molar specific activity as the substrate.
2. By a double isotope study using a mixture of the tritiated and, say, carbon-14 labeled compound. The ratio of the two isotopes administered is compared with the ratio in the metabolic products.

It is very important to have both labeled compounds of an identical radiochemical purity or a "spurious" isotope effect may be found [36]. Care must also be taken not to misinterpret a change in ratio due to an isotope effect for instability of the tritium label. So far isotope effects appear not to have proved a problem in such studies.

Concluding Remarks

This short review has attempted to illustrate some fundamental problems in the applications of tritium tracers in biological research, in particular the importance of using compounds with a known distribution of tritium atom(s). In addition, much more information is now available concerning the stability of the tritium atoms in molecules under various experimental conditions. The availability of this data and a recognition of the problems which are peculiar to this very versatile radioisotope of hydrogen adds confidence in the use of tritium compounds as tracers since the facts which are obtained are "true facts" and not "fiction."

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TRITIUM-LABELING
A REVIEW

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Tritium labeled compounds are widely used in many fields of study. The advantage of using tritium as a label lies in the low cost of the isotope, the ease of labeling, and the high specific radioactivity that can be achieved in the labeled material.

Tritiated compounds can be prepared with a specific radioactivity in the range of multicuries per millimole. Handling of curie amounts of tritium requires no radiation shielding for the operator, although inhalation or ingestion of tritiated compounds presents a health hazard, the magnitude of which depends upon their chemical nature and biological half-life. The short range of penetration of β-particles from tritium, especially in photographic emulsions, makes it the most desirable radionuclide for use in high resolution radioautography.

In contrast to carbon-14, tritium has a lower efficiency of detection, a tendency to become labile or undergo migration in enzymic and chemical reactions, and a large isotope effect. Incorporation of tritium into organics can be effected by nonsynthetic as well as synthetic methods. Antibiotics, plant alkaloids, and biopolymers which cannot be labeled with carbon-14 by synthesis, can be tritiated by nonsynthetic methods. A comparison of the characteristics of carbon-14 and tritium is given in Table 1.

There are a number of excellent books [1-3] and review articles [4-6] on tritium and tritium-labeling. This review will discuss mechanisms of several tritiation methods and their practical aspects. It will comment on the stability of tritium label in tritiated compounds, namely, protodetritiation, and briefly mention tritium hydrogen exchange for protein conformation studies.

Classification of Labeling Methods

Incorporation of tritium in organics can be achieved by nonsynthetic, synthetic, and biosynthetic methods. A sub-classification is given below.

- (A) Nonsynthetic methods based on:
    - (1) Self-irradiation - Wilzback labeling [9]
    - (2) Excitation and ionization induced by microwave, electrical discharge, γ-ray irradiation, Hg-photo-sensitization, etc., modifications of Wilzbach labeling.
    - (3) Recoil tritiums from nuclear reactions
    - (4) Exchange catalyzed by:

Table 1

Characteristics of Carbon-14 and Tritium

	^{3}H	^{14}C
$T_{1/2}$ (y)	12.26	5568
E_{max} (MeV)	0.0180	0.156
$\bar{E}_{\beta}$ (MeV)	0.0055	0.050
R_{max} (mg/cm^2)	0.54	29
$\bar{R}_{\beta}$ (mg/cm^2)	0.045	3.7
Resolution in radioautography (μ)	0.13-1.5	10-83
Recoil energy (eV)	1.65	6.9
Bonding orbital	s	sp, sp^2, sp^3
H-bonding, hyper-conjugation, etc.	yes	-
Primary kinetic isotope effect[a]	7.8	1.22
Activity per milli-atom (Ci)	29.1	0.062
Cost per Ci ($ US)	2	4500
Methods of incorporation	nonsynthetic, synthetic	synthetic

[a] Taken from Ref. (123).

        - (a) transition metals
        - (b) acids
        - (c) bases
    - (5) Radiation synthesis [7]
- (B) Synthetic methods:
    - (1) Hydrogenation
    - (2) Dehalogenation
    - (3) Reduction
    - (4) Tritioboration [8]
- (C) Biosynthetic methods:

In the following article, some of these methods for tritium labeling will be discussed.

Mechanism of Tritium-labeling

As synthetic and biosynthetic methods for tritium-labeling are inherently limited by the nature of chemical reactions and the functional capability of enzyme or cellular systems, the mechanism of reaction should not be unique towards tritium if the isotope effect is not considered. In nonsynthetic methods of tritium-labeling, incorporation of tritium occurs by substitution and exchange under the influence of a radiation field or catalyst. The efficiency of labeling is to a large degree affected by the nature and the physical state of the parent compound, the tritiating agent and the presence of additives. A knowledge of the mechanisms involved allows one to utilize the methods more efficiently for tritium-labeling and to perceive the relationship between various methods.

Wilzbach Labeling

In this method [9], the parent compound is exposed to several curies of tritium gas for days, followed by an extensive purification to produce the pure tritiated compound.

The mechanism of gas exposure labeling for homogeneous tritiation has been studied by Riesz and Wilzbach [10], Yang and Gant [11-14], Pratt and Wolfgang [15], Ache [16,17], and Cacace et al. [18,19]. They have investigated homogeneous systems containing tritium and one of the following hydrocarbons: methane, ethane, ethylene, propane, cyclopropane, n-hexane, and toluene. Isotope exchange in the methane tritium system has been studied by Wexler [20,21] using mass spectrometry and ion-cyclotron resonance spectroscopy. His results are in accord with those obtained by others.

Evidence from these gas phase studies leads to the postulation of the decay-induced and radiation-induced mechanisms in tritium-labeling. The mechanism is exemplified in the following with the extensively studied methane-tritium system [15,19,21].

In decay-induced labeling, the reacting specie is HeT^+ formed from the tritium molecule by nuclear transformation with a recoil energy of 1.65 eV.

$$T_2 \longrightarrow HeT^+ + e^- \qquad (1)$$

According to Wexler [22], the molecular ion, HeT^+, occurs to the extent of 94.5% among the decay products of tritium and has a lifetime greater than 10^{-5} sec in a collision-free space. This molecular ion, being a strong Bronsted acid, constitutes the principal tritiating agent particularly at low tritium concentrations when radiation-induced labeling is ineffective. It reacts exothermally with methane, according to the sequence of reactions:

$$\mathrm{HeT^+ + CH_4 \longrightarrow (CH_4T^+)^* + {}^3He} \qquad (2)$$

$$\mathrm{(CH_4T^+)^* \longrightarrow CH_2T^+ + H_2} \qquad (3)$$

$$\mathrm{CH_2T^+ + CH_4 \longrightarrow C_2H_4T^+ + H_2} \qquad (4)$$

$$\begin{tikzcd}[row sep=small] & \mathrm{CH_2T + CH_2} \\ \mathrm{C_2H_4T^+ + e^-} \arrow[ur] \arrow[dr] & \\ & \mathrm{CHT + CH_3} \end{tikzcd} \qquad (5)$$

$$\mathrm{CH_2T + CH_4 \longrightarrow CH_3T + CH_3} \qquad (6)$$

* indicates excited state

The ion-molecule $(CH_4T^+)^*$ is assumedly formed in the excited state and dissociates in approximately 10^{-11} sec. If it becomes stabilized by collisional deactivation within its short lifetime, the ion will react with the inactive methane by thermoneutral proton transfer to form tritiated methane:

$$\mathrm{CH_4T^+ + CH_4 \longrightarrow CH_3T + CH_5^+} \qquad (7)$$

Cacace _et al_. [19] found that $C_2H_4T^+$ is inactive toward CH_4 and in the presence of propane yields labeled ethane.

The efficiency of tritium molecules (T_2) and tritium hydride (HT) as tritiating agents, and the effect of oxygen in methane-tritium and toluene-tritium systems, have been compared [18,19], as shown in Table 2. Oxygen scavenges thermal radicals, but its presence has no effect on tritium-labeling of methane and toluene. This indicates that HeT^+ initiates the tritiation under the conditions given. The failure of HT which yields HeH^+ on β-decay to give any significant amount of the labeled product also emphasizes the role of HeT^+. That HeT^+ acts as an electrophilic agent in tritiation is further supported by the inductive effect manifested in the ratio of relative yield of tritiated products from trifluorotoluene, benzene and toluene (0.45 : 1 : 2.1) [18], and by the orientation effect exhibited in the considerably higher distribution of tritium in the _o_- and _p_-positions than in the _m_- and α-positions of the labeled toluene [16,18]. Kinetically, the rate of formation of the tritiated product by decay-induced labeling varies with the first power of tritium concentration [15].

In radiation-induced labeling, the β-radiation from tritium causes the formation of excited and ionized substrate as well as tritium molecules. The ionized molecules of the parent compound can give

Table 2

Effects of Oxygen on the Reactions of HeT^+ Ions with Methane and Toluene[a]

Tritiating Systems

System composition (mole %)	Activity, mCi		Yield, %		
	T_2	HT	CH_3T	C_2H_5T	C_2H_3T
Methane, CH_4	1.70	-	38	∿ 2	13
CH_4 (98) + O_2 (2)	1.70	-	40	∿ 2	13
CH_4 (98) + O_2 (2)	-	2.00	-	-	-
			(Toluene)	(Benzene)	(Methane)
Toluene, C_7H_8	1.70	-	49	2.7	∿ 8
C_7H_8 + O_2 (2)	1.70	-	60	3.8	-
C_7H_8 + O_2 (2)	-	1.70	(below detection limit)		

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[a] Taken from Refs. (18) and (19).

rise, upon charge neutralization, to radicals and intermediates which react with T_2, T atoms, or ion-molecules containing tritium to give the tritiated product. In the methane-tritium system the mechanism of reaction initiated by ionization of the methane component has been postulated to be as follows [15,19,21]:

$$CH_4 \rightsquigarrow CH_4^+ + e^- \tag{8}$$

$$CH_4^+ + CH_4 \longrightarrow CH_5^+ + CH_3 \tag{9}$$

$$CH_5^+ + T_2 \longrightarrow CH_4T^+ + HT \tag{10}$$

$$CH_4^+ + T_2 \longrightarrow CH_4T^+ + H \tag{11}$$

$$\hookrightarrow CH_3T \text{ precursor}$$

$$CH_4T^+ + CH_4 \longrightarrow CH_3T + CH_5^+ \tag{12}$$

For tritiation initiated by ionization of tritium in the mixture, the following sequence of ionic reactions were proposed [21]:

$$T_2^+ + CH_4 \longrightarrow CH_4T^+ + T \tag{13}$$

$$CH_4T^+ \longrightarrow CH_2T^+ + H_2 \tag{14}$$

$$CH_2T^+ + CH_4 \longrightarrow C_2H_4T^+ + H_2 \tag{4}$$

followed by reactions (5), (6).

The reaction of tritium and tritium ions with radiation-induced species of the parent compound leads respectively to 3/2 and 2-order terms in the kinetic expression of tritiation [15]. Based on scavenging studies with xenon and from an analysis of their rate equation for the formation of tritiated methane, Pratt and Wolfgang stated that radical-radical recombination, such as

$$CH_3 + T \longrightarrow CH_3T \tag{15}$$

is unlikely to be of importance in the labeling process. Their data yielded a linear log-log plot of the rate of formation of labeled methane vs. the tritium concentration [15].

For heterogeneous tritiation Wilzbach labeling constitutes a useful preparative source of tritiated compounds, but the tritiation procedures are carried out in heterogeneous systems with higher concentrations of tritium. The mechanism for tritiation of liquids and solids under such conditions is not entirely clear. No systematic studies similar to those cited above for gas-phase tritiation have been undertaken, although recent reports [23-30] on tritium-labeling in heterogeneous systems have furnished data from which a reasonable scheme of mechanisms may be deduced.

Tritiation in the heterogeneous phase requires a prior establishment of an adsorption-desorption equilibrium of tritium between the gas phase and the condensed phase on the walls of the container, followed by initiation of reactive species in the gas phase or at the interface [25]. The mechanism of gas phase initiation and interface initiation are given in Scheme I:

Scheme I:

Gas-phase initiation

$$T_2(g) \longrightarrow HeT^+ + e^-$$

$$e^- + T_2(g) \longrightarrow T_2^+ + 2e^-, \text{ etc.}$$

$$e^- + T_2(a) \longrightarrow T_2^+ + 2e^-, \text{ etc.}$$

$$e^- + M(\text{or wall}) \longrightarrow M^* + e^- \text{ or } M^+ + 2e^-$$

$$(HeT^+, T_2^+, \text{etc.}) + M \longrightarrow \text{tritiation}$$

Interface initiation

$$T_2(a) \longrightarrow HeT^+ + e^-$$

$$e^- + M \longrightarrow M^* + e^- \text{ or } M^+ + 2e^-$$

$$HeT^+ + M \longrightarrow \text{tritiation}$$

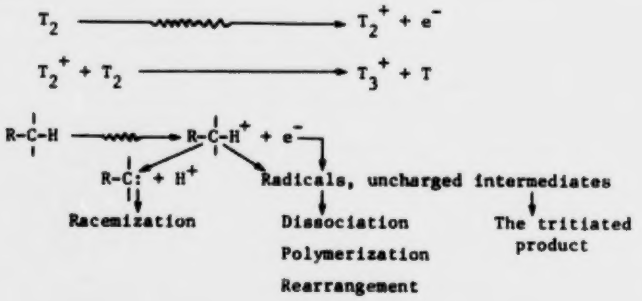
where $T_2(g)$ = tritium in gas phase, $T_2(a)$ = tritium in adsorbed phase, e^- = β-particle, primary or secondary electron, M = substrate molecule, and M^* = substrate molecule in the excited state.

The adsorption-desorption equilibrium of tritium between the gas phase and the condensed phase may serve as the basis for explaining the temperature [31], pressure [30,31], and surface effect [32] observed in gas-exposure labeling. At low temperature (77°K), when adsorption is favored, it is likely that the decay-induced labeling dominates; with increasing temperature the relative concentration of tritium in the gas phase increases and favors the radiation-induced labeling [25]. High pressures of tritium gas would shift the adsorption-desorption equilibrium at the interface, resulting in higher tritium concentration in the condensed phase and in increased labeling.

The surface effect in tritiation was observed by Wenzel et al. [32], who showed that when the substrate was adsorbed on charcoal and then exposed to tritium, the specific activity of the labeled product was increased 130-fold in the case of salicylic acid. The increase may be attributable to enhanced adsorption of tritium at the interface. When the exposure was carried out at -196°C, degradation of the labeled product was reduced [3], owing to a decrease in radiation-induced labeling. Meshi and Takahashi [33] found that the use of Pt-black or $Pt\text{-}PtO_2 \cdot H_2O$ catalyst intimately intermixed with salicylic acid for exposure to tritium increased the specific activity by approximately 1880-fold, as compared to that of the control. The Pt-black and $Pt-PtO_2 \cdot H_2O$ can chemisorb tritium onto their surfaces thereby promoting tritium exchange. When the catalyst was replaced with charcoal, no increase in specific activity was observed. Other metal catalysts have a smaller effect on the enhancement of tritium incorporation. The surface effect in the enhancement of tritium incorporation by charcoal has been observed in the Wilzbach labeling of L-tryptophane [34], but not in that of L-proline [34], or (-)-cocaine [35].

At high concentrations of tritium, the decay-induced labeling becomes less contributory to the total labeled yield, since the ion-molecule HeT^+ formed from β-decay of tritium molecule has a G(T) value (i.e., number of T atoms incorporated per 100 eV absorbed) of 0.018, or an L value (i.e., number of T atoms incorporated per T decay or 5700 eV absorbed) [12] of 1.0. The radiation-induced labeling involving ionic and radical mechanisms becomes dominant. It is likely that reactive molecular ions such as T_2^+ and T_3^+, besides T atoms and T_2 molecules, participate in the tritiation. A postulated mechanism of radiation-induced labeling is depicted in Scheme II [25].

Scheme II:



The scheme shows the process of tritiation involving ion-molecules containing tritium, tritium atoms, and radiation-induced reactive molecular species. The nature and abundance of radiation-induced reactive species vary with the tritium concentration and the chemical stability of the parent compound. Tritium labeling can occur by hydrogen abstraction, hydride transfer or radical mechanisms. In Wilzbach labeling, tritium can add to the -C=C- bond [36] by ionic and radical mechanisms [25] or replace I from -C-I bond [37] by a radical mechanism [23,24], leading to specifically labeled saturated or deiodinated product in an essentially carrier-free form. Radiolytic products, if formed with tritium incorporation, are usually of extremely high specific activity. The tritiation system is an exceedingly complex system from the viewpoint of radiation chemistry; one is constantly reminded of this by the difficulties encountered in purifying labeled compounds to achieve radiochemical purity.

The mechanism of tritiation can be modified by the introduction of different gases into the system. Oxygen [16,18,19,25], nitric oxide [12,15,16,30], iodine [15], iodine monochloride, and iodine monobromide [38] have been used as scavengers for thermal radicals. Oxygen inhibits tritium incorporation by radical mechanisms. It eliminates tritium substitution in the side chain of toluene but decreases the ring labeling only at high tritium concentrations (500 mCi) [16]. Tritium addition to oleate is also diminished by the presence of oxygen [25]. Rare gases have been added to the system with varying results. They act as moderators for charge transfer either to promote labeling or to inhibit it, depending upon their ionization potential and their relative concentration to tritium [39]. Thus, in systems containing high concentrations of tritium, argon [25,39] causes an increase in the yield of the labeled product, while in others, argon [16], helium [16,30], or xenon [15] decrease the degree of tritium substitution.

The tritiation system may be modified externally by means of microwave excitation [40-44], high frequency electric discharge [26, 45-47], γ-ray [45,48] and ultraviolet light [25,49,50] irradiation. The input energy from the external source will activate and dissociate tritium molecules and the parent compound into atoms, radicals and other reactive species so that they may combine to form tritiated products. Excited molecules and radicals are also formed by energy transfer from excited Hg ($^3P_1$) atoms by ultraviolet irradiation in photosensitization [49,50]. Tritium atoms can also be generated from heated tungsten filament [51]. The application of external energy to the tritiation system has the advantage of shortening the exposure time and reducing the quantity of tritium required for labeling. More recent modifications appear to favor an independent activation of the tritiating agent and the substrate in order to minimize substrate degradation. Protein radicals can be generated by γ-ray irradiation [48] or by electrical discharge [47] prior to exposure to tritiated hydrogen sulfide to yield the labeled protein (the free radical interceptor technique). Also, microwave activated tritium atoms can be directed onto a protein target outside the microwave region [52]. This technique requires only a very short reaction time and can yield labeled peptides and proteins of very high specific activity.

With respect to the distribution of tritium in the labeled compound, Garnett *et al*. [28] found tritium incorporation to be the highest at the most electronegative positions in tritiated anthranilic acid and isopropanol. The aromatic hydrogen is about an order of magnitude more easily substituted than the aliphatic hydrogen, and the secondary C-H bond is tritiated more efficiently than the primary C-H bond. Protic and ionizable hydrogens attached to electronegative atoms, O, N, S, etc., are efficiently exchanged. Tritium adds to unsaturated carbon-carbon bond [36] and replaced I from C-I bond [23,24,37]. Racemization occurs with amino acids and peptides [53,54] and with liquid octan-2-ol during exposure but the configuration is retained in the crystalline octyl phthalate esters [29]. It is likely that optical and geometrical configurations will be affected to some extent in most of the compounds during exposure [55]. In aromatic polycyclic hydrocarbons, no simple relationship exists between the efficiency of tritiation and the ionization potential [30]. The stability of the paramagnetic species of these hydrocarbons (anthracene > chrysene > biphenyl, naphthalene) in the presence of tritium is inversely related to their tritium incorporation [30]. Tritium labeling of the components in cyclohexane-benzene mixtures is determined by their electron fractions [17], indicating the absence of charge-transfer, but an enhanced tritium incorporation in phenanthrene in the presence of benzoic acid has also been reported [30].

The tritium distribution in toluene labeled by different methods is given by Ache *et al*. [26]; the radioactivity resides mainly (i) in the o- and p-positions by gas exposure, (ii) in the α-position by electric and microwave discharge, and (iii) in the o- and α-positions by mercury photosensitization. The difference in distribution is probably due to relative availability of tritium atoms and tritium molecular ions.

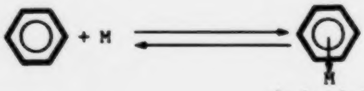
Recoil Tritiums from Nuclear Reactions

Tritium-labeling of organic compounds by recoil tritiums produced by the $^6Li(n,\alpha)T$ and the $^3He(n,p)T$ reactions has been reviewed by Wolf [56,57] and Wolfgang [58], and many others [59,60]. The method is of immense value for studying mechanisms of reactions of recoil tritium atoms. Owing to the inherent limitations placed on the experimental conditions, this method has not been developed into a general preparative source for tritiated compounds.

Exchange Labeling

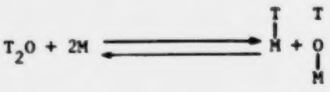
The tritium-hydrogen exchange reaction is frequently utilized to effect tritium labeling. The exchange of tritium between a tritiating agent and the parent compound can be brought about by the use of a transition metal catalyst, an acid, or a base at elevated temperatures. Many reactions that cause D-H exchange can be applied to T-H exchange if the deuterating agent is replaced with its tritiating analogue.

In this part, exchange effected by metal catalyst is considered. Two mechanisms of reactions, namely, the associative and the dissociative π-complex substitution mechanism, are postulated for T-H exchange between tritiated water and an aromatic molecule in the presence of a metal catalyst [61,62]. Their initial phase involves the formation of an adsorbed π-complex in which the organic molecule such as benzene lies horizontally to the surface of the catalyst. This is depicted as follows:



(π-bonded)

As rapid exchange occurs on the surface of the metal catalyst, the adsorbed tritium is furnished by the second reagent, tritiated water, by dissociative chemisorption, thus

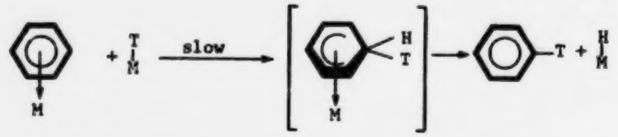


$$T_2O + 2M \rightleftharpoons \overset{\displaystyle T}{\overset{|}{M}} + \underset{\underset{\displaystyle M}{|}}{\overset{\displaystyle T}{\overset{|}{O}}}$$

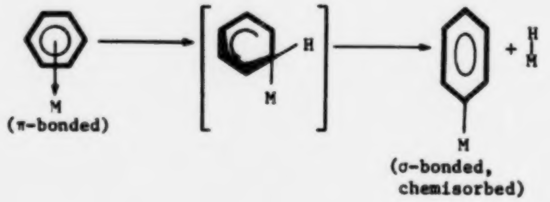
(chemisorbed)

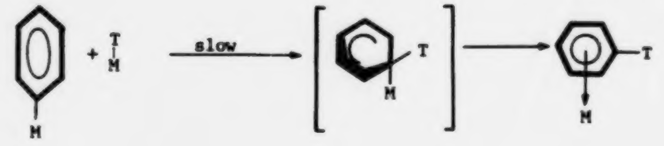
It may be pointed out that besides tritiated water, tritium and a tritiated acid such as acetic or pivalic [63], etc., can also provide the dissociative chemisorbed tritium atoms for exchange.

In the associative π-complex substitution mechanism, this flat π-complex will react with an adsorbed tritium atom at a nearby site on the catalyst by substitution to give the labeled compound.



In dissociative π-complex substitution mechanisms, the adsorbed aromatic molecule is rotated through 90° from its initial flat π-complex position, undergoes a π-σ bond transition at a 45° inclination to the surface of the catalyst and becomes σ-bonded in the vertical position by chemisorption.



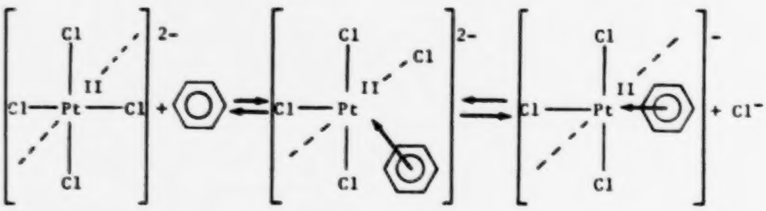


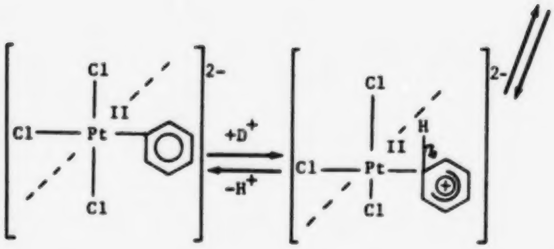
The σ-bonded complex can combine with an adsorbed tritium in its vicinity to give an intermediate which on return to the π-bonded state and desorption, yields the labeled molecule. The dissociative π-complex substitution mechanism has been found to be the predominant mechanism in aromatic exchange [64].

For efficiency and specificity, both the metal catalyst and the strength of the π-complex play important roles. Garnett et al. have studied the factors influencing the hydrogen isotope exchange in aromatics [64,65], heterocyclics [66,67], and aliphatics [68,69], using deuterium oxide as the second reagent. The formation of π-complex depends upon the ionization potential, electron affinity, and orbital symmetry of the organic adsorbate and the steric hindrance of its substituent group(s). A low ionization potential, a high electron affinity and symmetry of aromatic bonding and antibonding orbitals favor the π-complex formation. A competitive, strong π-complex adsorption occupying most of the active sites on the catalyst will retard the exchange by reagent displacement. Naphthalene, which is not efficiently labeled by the heterogeneous exchange method, belongs to this group. The π-complex adsorption is decreased by nuclear substitution in the aromatic molecule because of steric hindrance; the number and the bulkiness of the substituents determine the degree of inhibition of the exchange. Alkyl benzenes with bulky substituent groups, such as *tert*.-butyl benzene, 1,3,5-trimethyl-benzene, *m*-xylene, etc., are inefficiently labeled by this method.

The most effective catalyst for exchange in general is the platinum catalyst but elements from group VIII are effective. The metal catalyst may be prereduced with hydrogen, self-activated [71,72], or reduced with sodium borohydride [73]. The borohydride reduction is conveniently carried out at low temperature to avoid deactivation by thermal sintering. The self-activated catalyst is produced by reduction of the catalyst by organic compounds at an elevated temperature and is found to be more efficient than the prereduced catalyst for exchange in hydroxylated compounds [72]. Irradiations by ^{60}Co γ-rays and ultraviolet light [74] have an accelerated effect on the efficiency of exchange of the self-activated catalyst. β-radiation from curie amounts of tritiated water has a similar effect. This is attributed to the removal of chemisorbed oxygen layer on the surface of the catalyst by radiolysis prior to self-activation [74,75]. Elements of group VIII such as Ni, Ru, Rh, Pd, and Ir have variable catalytic effect and specificity. The isotopic exchange performed in the presence of nickel catalyst (borohydride reduced), showed a predominance of isotope incorporation in the aliphatic chain [73,76]. In catalytic exchange labeling of (-)inositol, it is possible to obtain labeled molecules with most of the tritium (92.5%) at C1 and C6 and with a minimum of racemization [72,75].

In contrast to *heterogeneous* catalysts for T-H exchange, Garnett and Hodges [76-79] introduced the *homogeneous* Pt catalyst for labeling of synthetic hormones and steroids [80] at a low temperature to minimize possible thermal catalytic degradation. The procedure requires heating in an evacuated sealed tube containing acetic acid, isotopic water, hydrochloric acid, the organic compound, and disodium platinum tetrachloride at a temperature within the range of 25-120°C until a given enrichment is attained. The acidity of the mixture is critical for maintaining the Pt catalyst in solution for the presence of any precipitated catalyst will cause the formation of by-products, such as chlorobenzene and biphenyl from benzene and result in exclusively acid exchange where this is possible [79]. The mechanism of homogeneous catalytic exchange involved a π-complex intermediate and can be depicted as follows:





The rate of exchange is directly proportional to the concentration of the Pt catalyst and benzene but varies inversely with the chloride concentration.

In homogeneous metal-catalyzed exchange, compounds such as nitrobenzene, which poisons the surface of the heterogeneous catalyst for rapid exchange, and naphthalene which binds strongly to the catalyst displacing the second reagent leading to inefficient exchange, can be labeled. At 80°C, the isotope exchange in naphthalene is exclusively at the β-position, whereas, at 130°C, random distribution results [79].

The π-complex adsorption mechanisms postulated for the aromatics can also be extended to heterocyclics and aliphatics [66,67]. Heterocyclics such as pyridines, quinolines, azines, picolines, lutidines, thiophenes, furans, etc., can be labeled by heterogeneous isotopic exchange. The mechanism involves a charge transfer adsorption of the heterocyclic compound onto the catalyst through the lone pair of electrons on the hetero atom, in addition to the π-complex adsorption. The combined effect is to orient the adsorbate molecule towards a favorable exchange at the α-position. Selectivity with respect to isotope distribution in the molecule depends upon the activation and the nature of the catalyst. Borohydride-reduced Co catalyst deuterates almost exclusively in the α-position of 2-picoline, whereas, the self-activated Ru catalyst causes exchange mainly in the 2-methyl group [67].

The aliphatics are rather inert towards catalytic exchange with the exception of those possessing unsaturation. Cholesterol is inefficiently labeled by catalytic exchange [4]. The presence of a carbon-carbon double bond in the molecule allows the formation of a π-complex involving an allylic group. The adsorption of a π-allylic system onto the catalyst has been postulated to be the mechanism of self-exchange of deuterium in propylene-3-d_1 [82] and of labeling of the methyl group in toluene [65]. Another mechanism postulated for isotope incorporation into aliphatics is the hydrogen abstraction reaction [65]. In tritium labeling of ethylene, the exchange with a labile carboxyl tritium in the presence of rhodium catalyst is postulated to involve an ethylene-rhodium complex [63].

Isotopic hydrogen exchange can also be induced between organic compounds and tritiating agents by Bronsted acids. The acid tritiating agents are: TCl [66,83], $TH_2PO_4 \cdot BF_3$ [84,85], ($AlCl_3$ + THO) [86-90], CF_3COOT [91], $THSO_4$ [92], $TClO_4$, TH_2PO_4, etc.

TCl The deuterated analog, DCl, was used by Calf *et al.* [66,83], to label cycloolefins. The mechanism involves the addition of TCl across the double bond followed by elimination of HCl to yield the labeled olefin. The conditions of labeling are mild, and the method has been applied to the isotopic hydrogen exchange in terpenes, and to the tritiation of thiamine hydrochloride [93]. Its usefulness for tritium labeling has probably been superseded by homogeneous catalytic exchange [83].

$TH_2PO_4 \cdot BF_3$ The phosphoric acid-boron trifluoride complex is a strong Bronsted acid and tritiates by electrophilic substitution. The distribution of tritium is mainly in the *o*- and *p*-position of the aromatic ring [85]. Electron donating substituents activate the ring and promote tritiation; their reactivity decreased in the order arranged: $-OH > -OCH_3 > -CH_3 > -SH > -SCH_3 > -OC_6H_5 > -H$. Electron withdrawing substituents such as -COOH, $-NO_2$, $-COOC_2H_5$, and -Br inhibit tritium at the *p*-position. Nitrogen-containing compounds cannot be tritiated with this reagent because of salt formation. Aliphatics are inert towards this reagent; in fact, cyclohexane has been used as the reaction solvent. According to Yavorsky and Gorin [84], the tritiated products are easy to purify with a minimum loss of radioactivity.

A modification of this method has been reported [94,95], employing a complex of tritiated acetic acid and boron trifluoride, for successful labeling of β-lipoprotein, ribonucleoproteide, somatotropin, insulin, dipeptides, and cholesterol. The substitution of phosphoric acid with acetic acid decreases the acidity of the complex, with a slight diminution in tritiation efficiency, but offers milder conditions, especially for tritiating biopolymers.

($THO + AlCl_3$) Tritiation can also be performed with tritiated water-promoted $AlCl_3$. At a molar ratio of 2:1 for $AlCl_3:H_2O$, an acid complex of the composition $T^+(Al_2Cl_3 \cdot OH)$ is formed [88]. It is a stronger Bronsted acid than $THPO_4 \cdot BF_3$ and affects tritiation by electrophilic substitution. The reagent has been used for labeling of aromatics, heterocyclics, peptides, pyrimidines and aliphatics. The specific activity of the tritiated aromatics and heterocyclics reflects their reactivity towards electrophilic substitution in the decreasing order: benzene > fluorobenzene > chlorobenzene > 2,4,6-collidine > 2,6-lutidine > pyridine > nitrobenzene [86].

This reagent has been applied for labeling uracil, adenine, diglycine and triglycine with a low efficiency of tritium uptake from tritiated water (0.2%) [88]. Cyclohexane and n-heptane are also labeled by the reagent, by a mechanism of hydride ion abstraction [89].

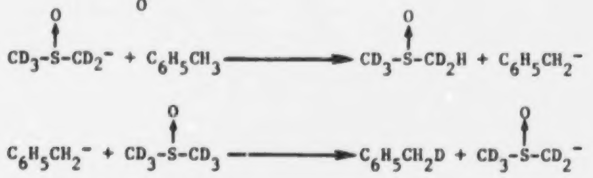
CF_3COOT This acid is frequently used for T-H exchange [91] because of its high solvent power and volatility and is preferred to other nonvolatile acids, such as $THSO_4$, $TClO_4$, TH_2PO_4, etc.

The exchange can also be base catalyzed. The rationale for base catalyzed exchange is that many C-H bonds in organic compounds have very feeble acidic properties and can dissociate in strong basic media to form carbanions. The basic ions utilized for T-H exchange are $-OH^-$,

$-OCH_3^-$, $-OC_2H_5^-$, $-OBut^-$ in tritiated water [96]. Other basic reagents used for exchange are basic alumina [97], silica gel [98], and tritiated ammonia [99].

(OH^- + DMSO) The hydroxyl ions in solution are hydrated; their catalytic ability is inhibited by the encirclement of water molecules. The addition of an aprotic solvent such as dimethyl sulfoxide (DMSO), dimethylformamide or tetramethylsulfone to an aqueous (OH^- $-H_2O$) medium decreases the solvation and increases the catalytic activity. The term "aprotic" is meant to indicate the solvent has no protons that can form hydrogen bonds. The alkoxide ions are stronger bases than the hydroxyl ion and can increase the rate of ionization. Their relative basicity, compared to OH^- in H_2O as unity increases in the order (OMe^- in MeOH)>(OEt^- in EtOH)>(But^- in t-BuOH) with a ratio of $3:20:10^3$.

Chen et al. [100], have deuterated toluene with deuterated DMSO in the presence of $CD_3-\underset{\overset{\downarrow}{O}}{S}-CD_2N$ a according to the reaction



$$CD_3-\overset{\overset{O}{\uparrow}}{S}-CD_2^- + C_6H_5CH_3 \longrightarrow CD_3-\overset{\overset{O}{\uparrow}}{S}-CD_2H + C_6H_5CH_2^-$$

$$C_6H_5CH_2^- + CD_3-\overset{\overset{O}{\uparrow}}{S}-CD_3 \longrightarrow C_6H_5CH_2D + CD_3-\overset{\overset{O}{\uparrow}}{S}-CD_2^-$$

The basic reagent of (DMSO + HTO) in the presence of OH^- or OBu^{t-} has been utilized to tritiate aromatics, alkanes, and cycloaklanes [96]. Tritium is incorporated exclusively in the alkyl chain of substituted benzene and randomly in benzene, naphthalene, n-heptane, and cyclohexane, but the distribution may be altered when the reaction temperature is increased.

The basic system for tritiation has several advantages, such as that DMSO is a good solvent for a large number of organic compounds. The labeled product is invariably obtained in pure form, tritium incorporation is usually achieved at room temperature, and no unsaturated compounds are hydrogenated.

Basic alumina, silica gel Basic alumina deactivated with tritiated water has been used by Klein and Knight [97] for labeling of keto-steroids by enolic exchange of tritium and hydrogen by liquid-solid adsorption chromatography. Silica gel treated with tritum gas is utilized by Nicolas and Paulaus [98] to tritiate ethylene, propylene, etc., by isotopic exchange. The efficiency of labeling of these methods is low in comparison with that of other catalytic exchange methods. Tritiated materials of high specific activity may not be obtained.

NH_2T Tritiated ammonia has been explored as a tritiating agent for exchange labeling of some nitrogen-containing compounds [99]. The exchange efficiency is very low, only asparagine and glutamine have specific activities above 1 mCi/mmole.

Practical Aspects of Tritium-labeling

The criteria for selection of a particular tritiation method will be dependent upon the specific activity and the radiochemical purity that can be achieved. The advantages and disadvantages of the principal tritiation methods are listed for comparison below.

Advantages of Wilzbach Labeling

1. Labeling can be readily carried out on a large body of organic compounds, natural products, and biopolymers with various concentrations of tritium at all temperatures. At -196°C, excessive radiation degradation is minimized.

2. The conditions of exposure are mild and can be applied to heat-sensitive compounds.

3. Tritiated compounds of high specific radioactivity can be obtained.

Disadvantages of Wilzbach Labeling

1. The effect of radiation is very substantial. The decay energy from 1 Ci of tritium is 1.8×10^{19} eV/day, which can lead to the disruption of a large number of chemical linkages in the substrate. The usual duration of exposure is from 7 to 14 days.

2. Labeled impurities are formed in an essentially carrier-free form, and their removal from the tritiated substrate requires extensive purification. The labeled impurities are produced by the radiation-induced mechanism, and their rate of formation is proportional to the 3/2 order term in tritium concentration. The labeled impurities may possess similar chromatographic behavior and crystal habit as the substrate so that separation by chromatography and crystallization becomes difficult. It is also likely that at trace concentrations the labeled impurities tend to concentrate at the interface and adsorb tenaciously to the surface of the substrate crystals.

3. In many cases labile tritium cannot be completely removed from the tritiated substrate and a constant specific activity cannot be achieved.

4. Unsaturated compounds are hydrogenated, and halogenated compounds are dehalogenated.

5. Racemization and isomerization may occur.

6. Position of the label in the tritiated molecule is unknown.

Application of Wilzbach Labeling

Numerous compounds, such as digitoxin [101], DL-glutamine [102], scopolamine [103], (-)-cocaine [35], azetidine, pyrrolidine, hexamethyleneimine, piperazine, methylcyclohexylamine, methylaniline, dimethylamine [104], sulfonylureas [105], phenyl benzene-thiosulphonate [106], amino acids and peptides [53,54] have been tritiated by the Wilzbach gas exposure method. Grosse et al. [107] labeled actinomycin by activation of tritium gas with a high frequency discharge. Gosztonyi, using microwave excitation in a modified Wilzbach method, labeled amino acids, local anesthetics [40], dihydrostreptomycin, tetracycline, antipyrine [42], purines, pyrimidines, and steroids [41]. Cipollini and Stöcklin [108] have compared the merit of tritium recoil and Wilzbach labeling of methylamine.

In spite of the shortcomings of the Wilzbach labeling, it provides the only means of introducing tritium labels into a heat-labile and/or complex biochemical molecule or biopolymer. Among the many modifications that were introducted to promote labeling and to retard radiation degradation, microwave excitation of only the tritium source appears to be a distinct improvement. Lieberman et al., have labeled the peptide, L-valyl-L-alanyl-L-alanyl-L-phenylalanine, and ACTH with a specific activity of about 2 Ci/mmole by this method [52].

Advantages of Catalytic Exchange

1. Due to the range of catalysts available, labeling conditions can be adjustable to the chemical nature of the substrate compound.
2. Labeling can be performed in heterogeneous or homogeneous condensed phase.
3. Tritiated products are usually recovered in very pure form and need only a minimum of purification to achieve radiochemical purity.

Disadvantages of Catalytic Exchange

1. Most of the exchange reactions require heating at 80-180°C for many hours to effect T-H exchange. Thus, it precludes the labeling of heat-sensitive compounds. Compounds that poison metal catalysts are also excluded.
2. Aliphatic compounds in general are not labeled with high efficiency.
3. Conditions of labeling may be such as to cause racemization and isomerization to occur, but to a lesser extent than those in Wilzbach labeling.
4. Detritiation may occur in the presence of a catalyst with higher acidity function than that used for tritiation.
5. The use of tritiated water and tritiated solvents poses a greater radiation hazard than the use of tritium gas.

Application of Catalytic Exchange

The exchange method has been widely used for tritiating a variety of organic compounds. The short reaction time minimizes the formation of radiation degradation products but radiation-induced exchange is a distinct possibility at high levels of tritium activity [109]. The exchange-out of tritium from the crude tritiated products during purification can be a substantial fraction of the total activity.

Advantages of Chemical Synthesis

1. Compounds are specifically labeled. The position of tritium in the molecule is definitely known.
2. Near carrier-free tritiated compounds can be prepared by tritio-hydrogenation and tritio-dehalogenation in one-step synthesis.
3. The tritiated products are obtained in radiochemically pure form.

Disadvantages of Chemical Synthesis

1. Synthetic procedures are often time-consuming and difficult. Frequently, existing procedures cannot be adapted to micro scale synthesis.
2. Many complex natural products and biopolymers are not amenable to tritium labeling by synthesis.
3. The specific activity of the final product is sometimes very much lower than that of the starting material.

4. Migration of tritium may recur in molecular rearrangement, isomerization, hydroxylation ("NIH Shift") [110], etc.

5. Multi-step synthesis offers a greater opportunity for personnel contamination and health hazards if intermediates of high specific activity are not cautiously handled.

Application of Chemical Synthesis

When the Wilzbach labeling and exchange labeling fail to yield a desired tritiated product of sufficient activity, one has to resort to a synthetic method to achieve the goal. Labeled derivatives of complex biopolymers can be prepared, such as tritiated acetylated derivatives of human growth hormone, bovine growth hormone, insulin [111], and trypsin [112], using acetic anhydride-^{3}H.

Catalytic dehalogenation with tritium gas is one of several preparative methods that lead to tritiated compounds of extremely high specific activity. The following compounds are examples: tritiated oxytocin [113,114], lysinevasopressin [114], DL-α-tocopherol [115] of high specific activity, specifically tritiated p-terphenyls [116] and biphenyls [117], chlorobenzene-3,5-^{3}H [118], phenylacetic acid-4-^{3}H [119], etc.

Advantages of Biosynthesis

1. The biosynthetic system is specific in nature and can yield a tritiated product from tritiated precursors with unimpaired biological activity.

2. Uniform labeling and specific labeling are possible.

Disadvantages of Biosynthesis

1. A satisfictory biosynthetic system is often not readily available.

2. Tritiated products of high specific activity are not usually obtainable because a high tritium concentration may cause radiation damage to the biosynthetic system.

3. Non-radioactive isotope dilution of the tritiated precursors may occur during biosynthesis.

4. Intramolecular migration of tritium is possible.

A large number of biosynthetic systems are given in the Proceedings of the Conference on Methods of Preparing and Storing Marked Molecules [121] and Labeled Compounds [122].

Stability of Tritium Label in Tritiated Compounds

The stability of a tritium label in tritiated compounds is of immense interest to those who use them for radiotracer experiments and to those who prepare them. Frequently, the merit of a tritiation method is evaluated on the basis of its ability to yield a stable product with respect to tritium exchange-out or protodetritiation upon purification.

The stability to protodetritiation is dependent upon bond linkages. Theoretically, tritium-containing bonds have lower zero point energy than hydrogen-containing bonds and will require slightly more energy to dissociate if anharmonicity is not considered. Experimentally observed maximum value of primary kinetic isotope effect for tritium is 7.8 [123]. As C-H bond is less ionic than -OH, $-NH_2$, and -SH bonds any hydrogen attached to a C atom in the molecule will presumably be stable, the only exceptions being α-hydrogens of carbonyl and carboxyl groups, secondary and tertiary hydrogens. Taking these facts into consideration, one is without an explanation for the demonstrated lability of tritium in the tritiated compounds.

The acidity of aromatic hydrocarbons has been studied by Eaborn and Taylor [124-126] and by Mares and Streitwieser [127], by detritiating substituted benzene and toluenes using $HClO_4$ or H_2SO_4 in aqueous CF_3COOH and lithium cyclohexylamide in cyclohexylamine (a strong Bronsted base). Parts of their results are given in Tables 3 and 4. In Table 3, as anticipated, the rates of protodetritiation of the substituted benzenes are higher in $HClO_4$ than in H_2SO_4 because the former is a stronger acid than the latter, but the rate ratio relative to benzene remains the same, indicating these compounds behave the same chemically in both media. In Table 4 are given the rates of dedeuteration and detritiation, the isotope effect, and the ratio of rates relative to toluene-α-d(t) of substituted toluenes with tritium label in the methyl group. The rates of detritiation are smaller than those of dedeuteration with an isotope effect of 3.3. Identical ratios of rates, relative to the labeled toluene, in both deuterated and tritiated compounds were observed, indicating again that the tritiated compounds behave chemically identical as deuterated compounds under the influence of the inductive and mesomeric effect of the substituent group.

If one accepts the protodetritiation as a manifestation of the lability of different C-H linkages as determined by their chemical reactivity, the phenomenon must indeed be universal. One may also conclude that a particular tritiated compound remains persistently impure, resisting efforts for purification, and that labile tritiums derive from tritiated impurities. To eliminate impurities as the source of labile tritiums, novel methods for purification by phase solubility and complex formation need to be emphasized. Purification of tritiated actinomycin by complexing with DNA [107] and of tritiated bilirubin with serum proteins [128], serve as useful examples.

To lend credence to the prevalence of hydrogen lability of C-H linkages one must examine the following facts: Hydrogen is univalent, forms covalent bonds by symmetrical s orbital, participates in hydrogen bonding and hyperconjugation, and ionizes as proton or hydride ion. The rates of the reactions are presumably controlled by many factors, slower in one medium than in others, and the ability to measure or detect the lability will be entirely dependent upon the sensitivity of the analytical method used. As radioactivity measurement is more sensitive than chemical analysis by a factor of 10^9 to 10^{10} and the smallest number of tritium atoms that can be readily measured is 10^5, the lability of hydrogens is more sensitively measured with tritium than by chemical means. Streitwieser's remark, "Titration during the course of a kinetic run showed no change in base concentration even when complete isotopic equilibration had been reached [129]," stresses the point.

In view of this inherent lability of hydrogens in organic compounds, it is possible to completely protodetritiate a molecule by exchange under rigorous and extreme conditions. The inference of this is that the stability of tritium in tritiated compounds is a meaningful one only on a relative basis. It is most likely that a tritiated compound when purified to satisfaction for one tracer experiment may not be suitable for another, if the conditions such as solvent, pH,

Table 3

Effects of Substituents on Rates of Detritiation of $X \cdot C_6H_4 \cdot T$[a]

X	H_2SO_4 medium[b]		$HClO_4$ medium[b]	
(Series A)	$10^7\ k_{ex}$[c]	f[d]	$10^7\ k_{ex}$	f[d]
H	3.6	1	14.3	1
m-Me	33	9.2	103	7.2
o-Me	1948	541	4720	330
p-Me	2526	702	4480	313
(Series B)[e]				
H	497	1	3025	1
p-F	860	1.73	5420	1.79
p-Cl	63	0.127	487	0.161
p-Br	36	0.072	298	0.098
p-I	43	0.0865	339	0.112

[a] Taken from Refs. (124) and (126).

[b] See original references for composition of the media and temperature of reaction.

[c] Experimental rates corrected for sulphonation.

[d] Ratios of rates, relative to benzene.

[e] Higher concentrations of acids used in this series.

Table 4

Rate Constants of Dedeuteration and Detritiation of $X \cdot C_6H_5CH_2D(T)$ at 25° Catalyzed by LiCHA[a]

	$10^5 k_{ex}$, sec^{-1}			k_{rel}[b]	
X	k_D	k_T	k_D/k_T	D	T
p-$(CH_3)_3Si$	17.5	5.90	3.0	4.70	4.57
o-F	44.6	15.5	2.88	(12)[c]	(12)[c]
m-$(CH_3)_3Si$	3.70	1.13	3.33	1.00	1.01
H	3.71	1.13	3.33	-	-

[a] Taken from Ref. (127). LiCHA = lithium cyclohexylamide in cyclohexylamine.

[b] Rates relative to toluene-α-d and -α-t, respectively.

[c] See A. Streitwieser, Jr., and H. F. Koch, J. Amer. Chem. Soc. **86**, 404 (1964).

ionic strength, etc., have been changed. Thus, in purifying tritiated compounds and applying them as tracers, joint criteria for the stability of tritium should be established.

Tritium-Hydrogen Exchange for Protein Conformation Studies

The tritium-hydrogen exchange between tritiated water and proteins has been ingeniously applied under a variety of conditions to study small differences in protein conformation. Kingman and Brisbin [130] investigated the rate and extent of exchange of collagen and tritiated water under different conditions and were able to distinguish four classes of hydrogen atoms. Emery [131] noted the number of slowly exchanging hydrogens in two cyclic hexapeptides (ferrichrome and ferrichrome A), their pH dependency, and conformational stabilization by iron. Hill and Leach [132] were able to detect the exchange between hydrogens at C-H cites and hydrogens from the aqueous medium under acid and alkaline conditions, and determined the extent of racemization and the labeling of amino acid residues in various proteins. Manning [133] has reported a method for determining D- and L-amino acid residues in peptides based on T-H exchange during acid hydrolysis.

Tritium-hydrogen exchange studies can also be used to demonstrate the disappearance of tightly bound, slow-exchanging hydrogens with increased radiation effect [134]. The hydrogens that are bound internally as N-H···O=C, in the helical portion of the polypeptide chain exchange slowly, those in the disordered portion exchange rapidly [135].

The binding of various metals to conalbumin affects both rapidly and slowly exchanging hydrogens [136]. The tritium-hydrogen exchange reaction has been advantageously used for *in vitro* labeling of DNA at high ionic strength by tritiated water [137]. The labeled DNA is the least damaged and retains complete biological activity.

ACKNOWLEDGMENT

A grant from the Academic Senate Committee on Research, University of California, San Francisco, is gratefully acknowledged.

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TRITIATION BY MICROWAVE DISCHARGE ACTIVATION OF TRITIUM GAS: A STUDY OF MECHANISM AND ITS APPLICATION TO THE LABELING OF ADRENOCORTICOTROPIC HORMONE (ACTH)

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This paper contains results dealing with the development of a method of general tritium exchange using microwave discharge activation of tritium gas, which is reproducible, reliable, inexpensive and applicable to most classes of organic compounds. The production of tritium labeled adrenocorticotropic hormone with a specific activity nearly 2000 times greater than previously available, with full biological activity is described. The investigation of some parameters of the labeling process in order to maximize the tritium-hydrogen exchange for further application to biologically important compounds is also discussed.

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Experimental Technique

The apparatus consists of a circular vacuum system equipped with a Raytheon Microwave generating unit (2450 MHz) and a unidirectional circulating pump. The target (reaction) area, which is several centimeters away from the microwave cavity, is a cold finger onto which the sample is placed. Use of Teflon stopcocks and a pressure transducer to measure the tritium pressure insures that the system is free of stopcock grease and mercury, since traces of these may interfere with the labeling.

Under normal operation, a few milligrams of sample are placed on the cold finger at 77°K. The system is then charged to a pressure of about 4 mm T_2 gas (<1 Ci) and the microwave power is turned on to generate a discharge plasma (red-purple in color) containing ionic and atomic tritium species which are swept toward the target.

The system as described is simple to construct, easy to operate and the results are reproducible over long periods of time. Unlike other methods (Wilzbach, electrical discharge, etc.), the geometry of the system and its energy requirements are such as to minimize substrate destruction.

Adrenocorticotropic Hormone

Since the report of Wilzbach in 1957 [1], many attempts have been made to apply this, as well as other modified gas exposure methods of nonsynthetic labeling, to the preparation of tritiated proteins and peptides. However, gross destruction of the protein, generation of multiple impurities and low specific activities have severely limited the biological usefulness of the techniques (Table 1).

Table 1

Tritium Labeling of Peptides

Compound	Tritiation Method	Sp. Act. μCi/mg	Tritium Incorporation # T atoms/# substrate molec.	Ref.
Insulin	Wilzbach	4.3	$1/1.2 \times 10^3$	2
Lysozyme Ribonuclease	Free Radical Interceptor (·,HTS)	0.7	$1/3 \times 10^3$	3
Lysozyme, actin	Interceptor/·,Elect. Discharge (HTS)	0.4-.06	$\sim 1/10^3$	4
Thyrotropin Releasing Hormone (TRH)	Wilzbach	8000	$\sim 1/10$	5
ACTH	Wilzbach	1.8	$1/4 \times 10^3$	6
Vasopressin	Electrical Discharge	400	$\sim 1/100$	10

In most cases (ACTH, insulin, lysozyme, etc.), statistically only one stable C-T bond was formed per every 1000 to 4000 substrate molecules, a very low incorporation indeed. Even in the very simple tripeptide TRH, only 10% of the molecules were labeled, which represents the greatest achievement for the simple Wilzbach labeling procedure.

In dramatic contrast to this, with our microwave activation of tritium gas technique, we have produced tritiated synthetic porcine ACTH at the very high level of 3.2 mCi/mg (14.5 Ci/mmol) with full biological activity. This represents a 50% statistical labeling (assuming only one tritium per ACTH molecule) and is nearly a 2000-fold increase over that previously reported by Nishizawa [6] in 1965.

Purification was carried out using a series of column chromatographic procedures (which are well characterized for ACTH), with final purity and homogeneity demonstrated by polyacrylamide gel electrophoresis showing a single peak corresponding identically with unlabeled ACTH.

The purified tritiated ACTH preparations were shown to bind to ACTH antibodies, give the same corticosterone response as unlabeled ACTH and were taken up by the adrenals of rats. In addition, the tritium uptake can be inhibited both by simultaneous injection of unlabeled ACTH, and by endogenous ACTH release. These facts well demonstrate that our purified tritiated ACTH retains both its immunologic and biologic activity.

Labeling Parameters

Due to the success of the ACTH labeling, which has demonstrated the feasibility of the microwave discharge technique, we have undertaken a more systematic and fundamental treatment of the labeling process. We hope to elucidate some of the parameters of the exchange, and possibly get some insight into the mechanism. For this study we have chosen an analogous series of aromatic hydrocarbons: 9-methylanthracene, xanthene and acridine. All the following investigations on parameter variation were carried out using 9-methylanthracene (9-MeAn).

Although the initial specific activity (unpurified 9-MeAn) increases with time, as shown in Table 2, the final specific activity

Table 2

Tritium Incorporation in 9-Methylanthracene as a Function of Time

# of exposures X time (minutes)	Rel. Sp. Act.
1 X 1 min	1
1 X 5	2
1 X 15	~2
3 X 5	2
1 X 90	~2

(after reaching constant specific activity) reaches a maximum between one to five minutes of exposure, after which time there is no significant change, even up to 90 minutes.

This implies that (1) some species, possibly partially reduced aromatics, are produced that compete favorably for the tritium and (2) short reaction times are desirable.

Runs made at 20, 50 and 100 watts of microwave power, P, show a linear increase of the initial specific activity with $P^{1/2}$, whereas the specific activity of the purified materials remains virtually unchanged as shown in Table 3. It is interesting to note here that in preliminary results of tritium atom concentration determination, using the hot platinum wire technique as described by Wood and Wise [7], a linear relationship between tritium atom concentration and $P^{1/2}$ is indicated.

Table 3

Tritium Incorporation as a Function of Microwave Power Input

Power Watts	Rel. Sp. Act. (Initial)	Rel. Sp. Act. (Final)
20	1.00	1.00
50	1.26	0.86
100	1.57	1.15

Table 4

Tritium Incorporation as a Function of Pressure

Pressure mm T_2	Rel. Sp. Act.
4	1.00
2+ 2 mm He	1.00
2	1.00
29	0.29

Table 5

Tritium Incorporation as a Function of Sample Weight

Weight mg	Rel. Wt.	Rel. Sp. Act.
4.300	11	1
0.382	1	6

Table 6

Tritiation of Various Organic Compounds

Compounds	Rel. Sp. Act.	Highest Carbon Free Valance[8,9]	Posit.
Naphthalene	1	0.452	1,4
9-Methylanthracene	19		
(Anthracene)*	(33)	0.520	9,10
Xanthene	47		
Acridine	78	0.542	9

* Tritiation carried out under slightly different reaction conditions.

Regarding pressure, as shown in Table 4, several observations are to be noted here: (1) Very high pressures (29 mm T_2) decrease the specific activity. This is possibly due to increased rate of recombination at high pressures. (2) A 50:50 mixture of He-T_2 gives results identical to that of an equal T2 pressure (4 mm). (3) At low pressures (2 mm), the final specific activity of the purified 9-methylanthracene is the same as at 4 mm but purification is more difficult. This may be due to the increase of tritiated byproducts caused from increased UV irradiation of the target area. In a microwave discharge plasma, a large UV flux is produced. As the pressure is lowered, the plasma grows, increasing the UV flux at the target, possibly giving rise to greater sample photolysis.

The most dramatic effect observed was from decreasing the sample weight as shown in Table 5. A decrease in weight, approximately a factor of 10, resulted in a sixfold increase in the final specific activity of 9-methylanthracene. This kind of increase was also observed with ACTH, especially when microgram quantities were used.

Reactions have also been carried out on related aromatic compounds. These results are shown in Table 6. The increase in specific activity going from naphthalene to acridine seems to be related to the increase in carbon-free valence, which is a measure of "residual bonding" ability, and is sometimes used as a measure of free radical reactivity. This may lend support to a free radical mechanism involving the attack of tritium atoms.

ACKNOWLEDGEMENT

Research was performed, in part, under the auspices of the U. S. Atomic Energy Commission.

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CANCER RADIOCHEMOTHERAPY WITH 7-TRITIOTETRACYCLINE:
I. RATIONALE AND PRELIMINARY ANIMAL RESULTS

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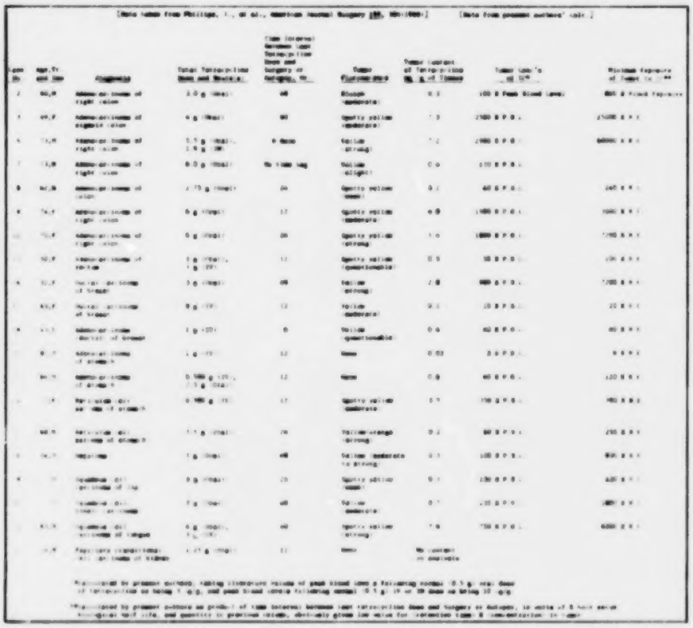
Rall et al. [1] first noticed that the broad-spectrum antibiotic tetracycline remained strongly associated with some varieties of neoplastic tissue in humans much longer than with normal tissues. Subsequent work indicated that the product of relative

Table 1

Tetracycline Uptake by Human Tumors



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retention time and relative tissue levels was often of the order of 100 to 1000, for a variety of human malignancies relative to normal tissues [2]. Results of some work of this type are shown in Table 1. Such results, though controverted by other investigators [3-9], have also received substantial corroboration in the literature [10-22,32]. Thus, the potential utility of tetracycline as a vehicle to selectively carry an antimetabolite to malignant growths seemed worthy of further study.

This paper presents a rationale for work using tetracycline, heavily labeled with tritium in the 7-carbon position, in malignancy treatment. Also presented are preliminary results of such a study in normal and malignant tumor-bearing rats. While this has been a small animal-number investigation, and results are not yet conclusive, it presently appears that this radiochemotherapeutic approach has yielded basically positive results in a conservative model system. Eventually, it may have therapeutic utility in high-risk cases of human malignancy.

Relevant Biological Properties of Tetracycline

Tetracycline has been recognized and clinically employed as an unusually effective broad-spectrum antibiotic since shortly after its discovery two decades ago. The tetracycline literature [23-25] is correspondingly vast, extending to well over 10,000 papers.

Tetracycline exerts bacteriostatic and, at higher concentrations, bactericidal action, primarily by inhibiting protein synthesis [26]. Tetracycline shows the following properties with regard to distribution, excretion and tissue binding. When injected intravenously, the antibiotic is rapidly distributed to most tissues and quickly comes to equilibrium concentration within and without the cell. The few exceptions are poorly vascularized tissue systems such as: large masses of necrotic or calcified soft tissue, hypovascularized tumors [2,32,33], bone which has been mineralized for some time [34], and the cerebrospinal fluid [25,67].

Tetracycline is fairly rapidly excreted, with most of the material being removed from the blood, essentially unaltered, by normal glomerular filtration in the kidneys. The initial serum concentration is cut in half after ~100 minutes in rats [26-29], and the corresponding biological half-life of tetracycline in humans is a little more than six hours [29,30]. Hepatic excretion accounts for 10 to 20% of the material removed [31]. Similarly, as the blood concentration of tetracycline falls, due to excretion, the intracellular and intercellular antibiotic levels fall with it [24,25,68]. Notable exceptions are newly mineralized bones [34,35] and, as will be discussed subsequently, certain malignant tissues.

It is well-known that tetracycline binds to normal mineralized tissues (e.g., bone, teeth) [28,35-37]. It is perhaps not as generally realized that this mineralized tissue binding is nonlinearly related to the peak blood level of the drug. Higher-than-therapeutic doses result in quite strong and persistent staining of the skeleton, while therapeutic or lower doses engender weak and transient labeling [39]. It also may not be generally appreciated that the most metabolically active few percent of the skeleton binds 99% of the complexed antibiotic [39,74]. This latter phenomenon is apparently associated with a much larger complex constant for tetracycline and freshly precipitated apatite than for the hydroxyapatite to which the microcrystalline apatite slowly hydrates under physiological conditions. Bone surfaces transiently exposed to tetracycline slowly lose their characteristic golden-yellow fluorescence over a period of days to weeks, depending on the peak tetracycline concentration and the ages of the various portions of the bone surface [39]. For instance, skeletally bound tetracycline in the mature rat is eluted with a time-dependence reasonably well-represented by two half-lives, one with a period of about ten hours, and the second of about eight days, each involving roughly equal amounts of complexed tetracycline [35]. The process in humans is semi-quantitatively similar [30,39,40]; most human skeletal masses bind tetracycline on their surfaces at a few times peak blood concentrations for about a soft tissue biological half-life, while the "hot spots" on the skeleton bind it at several dozen times peak blood level for longer periods [39].

As mentioned earlier, tetracycline localization and retention in human malignancies was first noted 14 years ago [1], when breast cancer tissue examined at autopsy was observed to fluoresce yellow-green under ultraviolet light. The fluorescence in the neoplastic tissue was determined to be due to the presence of tetracycline administered several weeks before autopsy. Many subsequent studies established tetracycline fluorescence as a highly reliable and specific diagnostic tool for malignancy screening [51,52]. For instance, it has come into general use for detecting gastric carcinoma [6,21,52-58] and other types of human tumors [15,21,59-62].

The strong tetracycline fluorescence often observed in malignancies [2,10-20] is apparently associated with unusually high concentrations of calcium ion in such tissues [42], as well as with greatly increased levels of the antibiotic [2,43]. Indeed, tetracycline localization in certain malignant cell types (e.g., von Kossa-positive cells) is apparently associated with formation of hydroxyapatite-like microcrystallites in the cellular cytoplasm during their actively metabolizing and high mitotic index periods [3,16,22,42]. Since we are considering tetracycline as an antimetabolite vehicle, only the time-dependence of its absolute concentration is of interest. Thus the fluorescent intensity is not an accurate gauge of concentration. Relatively great weight must be placed on semiquantitative, absolute analysis techniques [2,26,33,43].

If tetracycline is to be used to carry and concentrate an antimetabolite into malignant tissues, at least two factors need to be taken into consideration. First, is the antibiotic localized within the cells, or extracellularly? Second, is the tetracycline deposited in the actively growing or latent, possibly necrotic, portions of the tumor tissue? There is a large body of somewhat conflicting evidence in the literature bearing on these questions [2-20,22]. It is the opinion of the present authors that the preponderance of the evidence in the literature, corroborated by data from our present experiments, favors (a) intracellular localization of tetracycline and (b) localization within the actively metabolizing neoplastic cells [2,12-20,22].

Additional questions arise in the choice of a specific antimetabolite. Ideally, one would like an antimetabolite which has a combination of properties for maximum therapeutic effect. Specifically, it should minimally effect normal tissue. It should be deposited in a location within the malignant cell where it has maximum probability of disrupting the cell's functions. It should be effective in all those tumors which show significant tetracycline binding, and should not interfere with that tetracycline binding. It should be very firmly attached to the carrier molecule so as to minimize damage to normal tissues through dissociation into tetracycline and an untargeted antimetabolite, and to maximize the likelihood of fatally injuring the tetracycline-accumulating cell. Once in the target cell, it should have a high probability of injuring that cell in low enough administered doses to avoid possible tetracycline toxicity to the entire organism.

Several groups of investigators have considered these matters previously, and evaluated their choices experimentally. Ayres [44] first employed the 7-chloro derivative of tetracycline (known clinically as Aureomycin), as well as tetracycline itself [12], topically and systemically to treat invasive forms of cervical cancer. Recession and often apparently complete extirpation of a majority of the anaplastic lesions were observed during the treatments. However, in most of the cases, the neoplasms recovered and continued to proliferate when this therapy was discontinued. An adduct of nitrogen mustard, a well-known antimetabolite long employed as an antimalignancy drug, with tetracycline was apparently the first tetracycline-based material to be evaluated as an antimalignancy agent [45]. Results were deemed mildly positive by the investigators, in spite of extensive chemical decomposition of the adduct under physiological conditions. Compounds containing ^{131}I, an eight day half-life β^- and γ-ray emitter, were complexed to tetracycline and studied in the treatment of mouse malignancies by other groups [46-49]. Seemingly quite positive results were obtained, including several-fold life prolongations, and tumor remission and necrosis induction in several cases. Unfortunately, the ^{131}I tended to decomplex and home on the thyroid. Other complications were also involved.

Considering this history, substitution of a radionuclide on the tetracycline molecule in place of a stable nuclide of the same chemical properties seemed capable of meeting most of the above requirements. Specifically, if the radioisotope were an α- or β-particle emitter, the biological damage could be quite localized and very intense, relative to a chemical antimetabolite. Specific damage could be quite high if the radionuclide half-life were not overly long, and the selectivity of the administered radiation dose could be great if the radioactive half-life were greater than the biological half-life. Radionuclide decay products would be severely damaging to all cells for which tetracycline showed a sufficiently strong affinity. Also, substitution of a radioactive, but chemically nearly identical, atom would suffice to insure pharmacologically identical behavior of the tetracycline. Affixing the radionuclide in question to one or more of the carbon atoms of the backbone of the molecule would almost guarantee that it would remain attached essentially indefinitely under physiological conditions, considering the extraordinary in vivo chemical stability of tetracycline [24,26,27]. Finally, extremely large radioactive doses could be administered without reaching the tetracycline toxicity dose regime.

Consideration of these factors and chemical synthetic feasibility strongly suggested that tritium was the radionuclide of choice. The biological half-life of tetracycline in normal, nonmineralized human tissues is about six hours, so that the 12.3 year radiological half-life of tritium is much greater than the biological time scales for tritiotetracycline retention, even in malignant tissues. The very soft tritium decay betas have path lengths in biological tissue of subcellular scale lengths. Thus, they very satisfactorily meet the "localized kill" requirement, and they give relatively satisfactory bone surface dose behavior.

It should be recalled that low energy beta rays in general, and tritium betas in particular, have very short ranges in moderate Z materials, such as bone. Characteristic path lengths of tritium betas are only a few thousand Angstroms in bone, and about 10^4 Å (1μ) in soft tissue. Thus, only tritium atoms within a few tenths of a micron of bone surfaces will decay to dose tissue of interest, such as the bone-metabolizing cells. Bone itself is effectively totally immune to radiation damage in the dose regimes of interest. Loading of freshly mineralized bone surfaces by labeled tetracycline may be suppressed by such means as pre-saturating the active sites on the skeleton with unlabeled tetracycline. Parathyroid hormone treatment 12 to 24 hours prior to labeled tetracycline treatment may also be of use. This latter procedure secures the desired result somewhat indirectly, by stimulating osteoclastic activity and slowly ablating away the skeletal surface which has absorbed the radioactive antibiotic. Both of these procedures have some disadvantages and intrinsic complications [41], and may be unwarranted, particularly if little of the skeletal surface is freshly mineralized, as in adult individuals. It should be realized that the metabolically active skeletal "hot spots" appear to lay down apatite at the rates in excess of 1 μ/day [39]; such deposition rates would suffice to completely "bury" tritiotetracycline within about a soft tissue biological half-life (six to eight hours) after it was apatite-bound, as far as its radiobiological significance is concerned. For these reasons, 7-tritiotetracycline was chosen as the labeled tetracycline analog for experimental evaluation of the malignancy radiochemotherapeutic procedure discussed above.

It is useful at this point to consider the dose and dose rates available from 7-tritiotetracycline. Typical multiple-time intravenous injections of tetracycline for antimicrobial therapeutic purposes are about 10^{-5} grams of tetracycline per gram of organism [24,25]. This dose will therefore be taken as a reasonable one, biochemically speaking. Tetracycline seems to distribute about the organism, and equilibrate with both intercellular and intracellular fluid, in a few minutes [24-26]. Thus, a completely labeled 7-tritiotetracycline dose (containing about 50 Ci of tritium) would deliver a peak dose rate of about 10 rads/h to average soft tissues. Taking the biological half-lives of tetracycline into account, this would amount to a total time-integrated dose of about 20 rads in rats, and of about 60 rads in humans. Naturally, the increased relative concentration of tetracycline in tumors would result in increased relative doses to these areas. Previous investigators [2] have found the concentration of tetracycline in some human neoplasms to be larger by a factor of 10^2 to 10^3 than that in normal tissue, and to be retained more than ten times longer, relative to normal tissue. If 7-tritiotetracycline is similarly concentrated and retained in malignant tissue, the above-mentioned doses to normal soft tissues would result in a dose of several kilorads to several tens of kilorads to the malignant tissue. The corresponding dose rates would be of the order of a kilorad/h. These dosage levels are, of course, sufficient to kill all mammalian cell types, especially high mitotic index, neoplastic ones [50].

The question of relative biological effectiveness of 7-tritiotetracycline is a relevant one, since tritium-decay betas have path lengths considerably less than a typical cell diameter and they deposit energy quite nonuniformly along their paths [63]. An assumed RBE equal to 1 (i.e., of the same degree of radiobiological effectiveness as tritiated water) has not yet been proven. Several investigators have reported observations of tetracycline localization within cells

[12-20], particularly at or near the extremely radiosensitive nucleus [12,18]. Our semi-qualitative observations to date have merely established that, in rats, the RBE is certainly less than or equal to about 30, and probably less than or of the order of 10. This was deduced from the lack of radiation sickness effects when 10 to 15 rad dose equivalents of the labeled antibiotic were administered. Studies of intracellular localization of tetracycline have been relatively rare particularly with respect to human malignancies. In the absence of conclusive results from our own work at present, this aspect of the rationale for our radiochemotherapeutic approach must be said to rest on a quantitatively insufficiently secured foundation.

The factors mentioned in the foregoing discussion were deemed sufficient motivation to undertake studies of 7-tritiotetracycline metabolism and binding in normal and tumor-bearing animals, specifically rats. An account of a modestly scoped experimental study along these lines, still underway, constitutes the remainder of this paper.

Experimental Studies with Non-tumor-bearing Rats

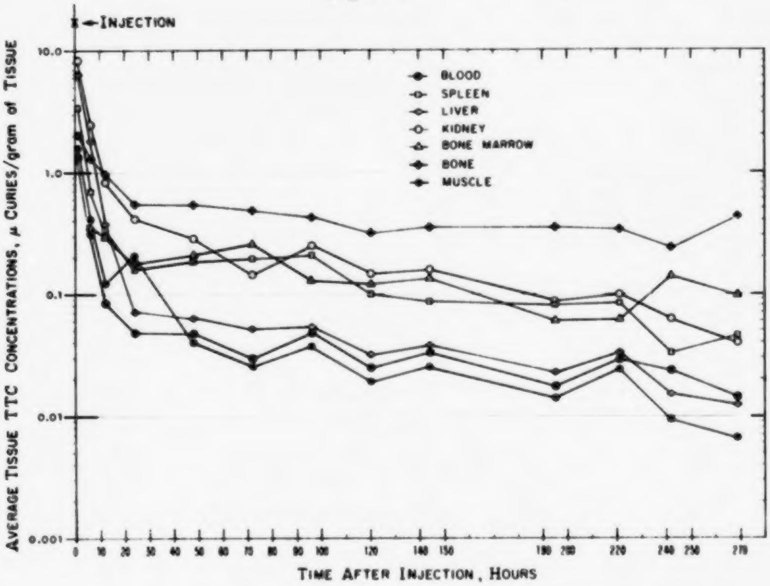
The fundamental purposes of the experimental studies with normal (i.e., non-tumor-bearing) rats were to verify and extend previous tetracycline metabolism studies in a variety of animals, to investigate its binding to mineralized tissues, to study its possible intracellular localization and explore the related implications for the RBE of tetracycline-bound tritium.

The 7-tritiotetracycline (TTC) for these studies, as well as for those with tumor-bearing animals, was prepared from reagent 7-chlortetracycline by catalytic reduction in an excess of tritium gas, in a variant of a commercial procedure [64], and was obtained in 90+% yield. Following washing off of loosely bound tritium with warm acid, the crude TTC was purified by a cycled combination of tetracycline characterization procedures [65], and superquick-frozen and stored at liquid nitrogen temperatures [66], often in the presence of radioprotective chemicals [66], to minimize radiochemical damage. Product analysis was by paper chromatography and liquid scintillation counting.

A suitable biological analytic procedure was found, in the form of forced complete oxidation of tissue samples, and subsequent scintillation counting of the water condensed from the combustion train exhaust. Sample combustion was accomplished in a Packard Tri-Carb 300 Tritium Oxidizer. Inter-sample system flushing was used to reduce memory in the oxidizer unit to the 0.1% level, as verified by repeated blanks. Samples were counted in a Nuclear Chicago Mark I liquid scintillation counter.

Normal (non-tumor-bearing) animal work was initiated to establish retention times of TTC in different rat tissues. Sprague-Dawley and Wistar females were used, with groups of animals (generally 15 to 20) injected intravenously (jugular cut-down) with specific quantities of activity (usually in the range of 1 to 30 mC). Commencing one hour after injection and continuing at three hour intervals for the first 24 hours, and once every 24 hours thereafter, an animal was sacrificed. Sample tissues from liver, kidney, spleen, femoral muscle, bone, bone marrow, blood, feces and urine were removed for analysis and frozen until processed. Early in the study, lung, heart, ovary and thyroid tissues were also collected. Collection of these tissues was subsequently discontinued, as they were always found to have relatively low TTC concentrations, which were proportional to those of liver and kidney. Triplicate samples of 150 mg were taken from those tissues and oxidized

Figure 1



and counted. The resulting tissue levels of TTC as a function of time from one such study, involving 18-month-old Sprague-Dawley females, are indicated in Figure 1.

Studies of procedures involving pre-dosing with unlabeled TC in order to "pre-load" mineralized tissues (though the necessity of this pre-loading has yet to be established) have been initiated, though no definite results are available at this time. Additionally LD_{50} tests on normal animals indicate the TTC radiotoxicity levels to be grossly higher than those which we find to be therapeutically effective in tumor-bearing animals.

Autoradiographic studies have also been undertaken to attempt to establish and semi-quantify the intercellular and intracellular localization sites of TTC in various normal and, subsequently, malignant tissues. We also hope to improve the mechanistic understanding of TTC radiation damage of such tissues and, in particular, to provide microscopic data to correlate with macroscopic observations of the RBE of TTC, such as might be obtained by gross indications of 100 to 500 rem doses delivered to animals. The solubility of TTC in tissue and the rapidity of small-scale diffusive transport rendered standard autoradiographic procedures imperfect. We therefore do not consider our present results conclusive, even for normal tissues. However, data developed to datedo not indicate strong localization of TTC in or about any intracellular structure in rat liver. Some mitochondrial localization of TTC in liver has been reported previously [19]. Nor did we find any significant extracellular concentration sites. This of course would imply near-unity RBE for TTC, at least in rat liver. These studies are continuing.

Tumor-bearing Animal Studies

Our tumor-bearing animals come to us from a laboratory-animal breeding colony which produces Sprague-Dawley experimental animals. Some significant fraction of colony female breeders spontaneously develop malignant tumors in one or more of the mammary glands sometime during their breeding lifetime. Rat mammary tumors constitute a rather conservative model system for purposes of our study, inasmuch as they have been reported to have a relatively low affinity for tetracycline [3]. These animals have come to us bearing tumors of masses between a few grams to over 100 grams. Spontaneously arising tumors were deemed interesting because they were thought to be more similar to those of human malignancies than transplanted neoplastic cell lines. Our concern was that relatively subtle metabolic or antigenic properties of these latter lines may have altered significantly during multiple animal passage.

Upon arrival, the animals have been examined and, by tumor size and animal health, allocated to one of several study programs. Animals with excessively large tumors (>60 grams) have been considered as most suitable for relatively short-term TTC localization studies. Animals with tumors of small to intermediate sizes have been assigned randomly to various programs, such as long-term localization and retention tests, radiochemical and biochemical toxicity studies, unlabeled TC and tritiated water control experiments, therapeutic dosage regimes, and several others. From each of these tumor-size groups, numerous animals have been used as uninjected controls to study the development of these tumors, since their spontaneous nature renders them rather variable in microscopic and macroscopic appearance and behavior.

Injections for tumor-bearing animals were done intravenously, as with the normal animal studies, and animals were frequently maintained thereafter in metabolic cages, to facilitate urine and feces collection during the relatively crucial several hours after injection. At sacrifice, animal tissues were excised and processed according to the procedures outlined for normal animals. The tumor(s) was (were) removed with considerable care, due to the desirability of excising essentially all the malignant tissue; this was generally accomplished with relative ease, because of their usually encapsulated natures. Those which had invaded surrounding tissue were removed with the intentional inclusion of some normal tissue. This latter tissue was easily identifiable as such during analytic procedure because of its grossly lower TTC content, as well as by appearance. As with normal animals, triplicate 150 mg tissue samples were oxidized and counted. However, because of variability in gross animal weight and animal mass vs. tumor mass, but more importantly, because of the relative scarcity of the animals themselves, we do not as yet have complete data sets indicating TTC levels as a function of time, tissue type, administered dose, age and general condition for the tumor-bearing animals. Also, our large sample mass analytic procedure is not well-suited to high spatial resolution studies. What we did find, though, were tissue levels of TTC in the average tumor-bearing animal's normal tissues corresponding well to their counterparts in healthy animals, taking into consideration time after injection and administered dose. These "normal tissue" levels, however, were somewhat lower than in non-tumor-bearing animals because of the substantial amounts of TTC bound in the tumor. Indeed, it appears that the mammary tumors absorbed and bound a substantial fraction of the injected TTC in the first millimeter or so of surface tissue layers, and in annular regions immediately surrounding blood vessels penetrating the tumors. Our continuing studies indicate that TTC was not strongly bound in the grossly necrotic areas of the tumor. Since all tumor portions were available for and subjected to identical work-up, this statement is based on absolute amounts of tritium (presumably ^3H bound to tetracycline, as noted before [26]). Previous fluorometric indications of TC concentrations are known [42] to be quite significantly dependent local Ca^{++} concentrations.

However, of more ultimate significance are the therapeutic findings. In animals with initial tumors of masses under about 7% of body weight, apparently complete regression of tumors with no discernible ill effects to the overall organism may be fairly uniformly obtained; five of the seven animals so treated were maintained in good health six months to one year after final injection. In cases of animals with initial tumor masses between 10% and 20% of body mass, TTC treatment apparently results in arrest and occasional regression of tumor growth. In 10 animals of 14 so treated the animals survived in apparently good health for six months to one year. Control animals in the former group became moribund with ≥100 g tumors and were sacrificed after about three months, while animals in the latter group usually required sacrifice within six weeks. As mentioned earlier, dose schedules have varied from animal to animal, as several approaches have been simultaneously explored, and control work remains difficult with this type of spontaneous tumor and with our tumor-bearing animal acquisition opportunities. We can tentatively conclude from our control work that there are essentially no spontaneous remissions for this type of tumor, for we have observed no case of tumor regression in a total of 31 untreated animals. These results are summarized in Table 2. We are continuing attempts to determine the reason(s) for incomplete success of TTC therapy.

Table 2

Results of the TTC Therapy on Rat Mammary Tumors

Stage	Qualitative Results	Quantitative Results
Initial tumor mass (4-20 g) ≲7% of body mass (250-325g); several TTC injections	Partial to total tumor regression; 6-12 month survival periods to date	5 animals of 7 treated
Initial tumor mass(1-20g) ≲7% of body mass (250-325g); no injections (CONTROL)	Required sacrifice in ~3 months	5 animals of 5 studied
Initial tumor mass(25-60 g) ≳10% and ≲20% of body mass 300-400g); several TTC Injections	Tumors in stasis or regressing; 6-12 month survival periods to date	10 animals of 14 treated
Initial tumor mass (25-60g) ≳10% and ≲20% of body mass (330 -400 g); no injections (CONTROL)	Required sacrifice in ≲2 months	8 animals of 8 studied

Implications of Study Results to Date

Such implications as may be drawn regarding the possible efficacy of labeled tetracycline in human malignancy radiochemotherapy derive almost exclusively from the work of previous investigators at this time. That ionizing radiation doses in the kilorad regime are very effective in treating malignant growths has been well-known. Indeed, the goal of cancer radiotherapy has been to dose malignant growths in an individual at such levels, without so dosing much normal tissue. Many ingenious schemes have been developed to this end. It has been well-established over the last decade that the antibiotic tetracycline was strongly concentrated and retained in at least some varieties of human and animal malignancies. It was demonstrated in the present work that a highly radioactive and chemically identical analog of tetracycline, 7-tritiotetracycline, could be synthesized, purified and stably stored (three to six months) in concentrations of experimental utility. The calculational results presented in the foregoing indicated that metabolically acceptable levels of 7-tritiotetracycline (of the order of 10 mg/kg of body weight) could apparently engender whole body doses of several tens of rads, while delivering localized doses of the order

Table 3

Some Possible Failure Modalities of Tetracycline Malignancy Radiochemotherapy

Modality	Likelihood
Tetracycline is not strongly absorbed (or retained) by some significant human malignancies.	Negligible
Tetracycline, though absorbed strongly by some significant human tumors in bulk, is confined to necrotic portions of all such tumors.	Negligible
Tetracycline, though absorbed strongly by actively metabolizing portions of some significant human tumors, is not found intracellularly.	Negligible
Tetracycline, though absorbed strongly and intracellularly by actively metabolizing portions of some significant human tumors, is not so absorbed by all neoplastic cells therein.	Small → Moderate
Tetracycline, though absorbed strongly and intracellulary by all actively metabolizing cells in some significant human tumors, is also strongly and intractably absorbed by some essential normal tissue, e.g., bone, which would be radiation-dosed to an unacceptable extent.	Small → Moderate

of ten to tens of kilorads in some tumors. These facts might reasonably lead one to the conclusion that 7-tritiotetracycline might be usefully employed as a radiochemotherapeutic agent in the treatment of the susceptible varieties of human and animal malignancies.

We wish to suggest no more than this, and to point to the somewhat positive malignancy suppression results obtained by us. It is suggested thereby that TTC may be therapeutically useful. The obvious conclusion is that the question of its utility should be resolved, and it is toward this end that our efforts are directed. It should be understood, however, that results of this study are preliminary, and subject to revision as data sets become more complete. Many essential portions of this investigation are still mid-course, and very few conclusions may be unequivocally drawn at this point in time. Some of the more obvious failure modalities of this scheme, and our subjective assessments of their likelihood, are indicated in Table 3.

Somewhat more generally, it should be pointed out that tetracycline is certainly not unique in its affinity for certain types of malignant growths. Other compounds [69-73] likewise home in on neoplastic cells. If these compounds are of relatively modest molecular weights (e.g., $\lesssim 10^4$ atomic units), and if tritium may be added to the molecule at at least one site without perturbing its pharmacodymanics significantly, then they may be quite useful as anticancer agents. Tritium, because of its intermediate length half-life, its unique decay mode into very soft, subcellular path length beta rays, its economic availability in a wide variety of high specific activity chemicals, and the ubiquity of hydrogen in biochemicals, seems likely to be a uniquely valuable constituent of such approaches.

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TRITIUM HYDROLOGY STUDIES IN CALIFORNIA

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Tritium is an ideal tracer for water since it is part of the water molecule and follows the complete hydrologic cycle. This makes it a useful medium for measuring the processes that transport water. Its 12.4 year half-life provides a method for determining the time since the water was in the atmosphere. The first measurements [1] were used as exact age determinations after the production rate by cosmic rays was established. Later measurements [2-5] made use of the tritium fallout peaks, which were produced by thermonuclear weapons tests, to determine the mixing ratio or amount of recent water added to a reservoir. With the exception of the deep ocean and certain ground waters, the large amount of thermonuclear tritium (produced mainly in 1954, 1958, 1961, and 1962) masks the cosmic ray tritium. Other laboratories have also measured the thermonuclear fallout tritium [6]and the application of tritium to hydrology has been used in several areas [7-11].

The UCLA Tritium Laboratory has been engaged in a number of projects that were not reported in our earlier publications, which included hydrology [3,12,13]. Brief accounts of some projects have appeared in our reports [14], and a few of the data have been given in the International Atomic Energy Agency lists [15]. The results are given in Tritium Units (TU); one TU is a concentration of one tritium atom to 10^{18} hydrogen atoms.

Experimental

The analysis technique has been described [1] and includes three or four electrolysis steps (to enrich the tritium by a factor of 400 to 1000), deuterium analysis to check enrichment, and geiger counting using sample hydrogen with argon and ethylene. From measurement of standards, blanks, and replicates, the overall precision is about ±10%. For samples very low in tritium, the errors are given in Figure 3.

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Los Angeles County Artificial Recharge

In 1961, in cooperation with the Los Angeles County Flood Control District, a project was initiated to measure the tritium in wells in the section of Los Angeles known as the Downey Plain (approximately 15 km north of Long Beach and 15 km southeast of the Los Angeles Civic Center). The area of the study, Figure 1, includes an artificial recharge area (Rio Hondo and San Gabriel Spreading Grounds) which receives imported water via the Metropolitan Water District Aqueduct from the Colorado River. The basin sediments are chiefly coalescing alluvial fans of the Los Angeles, San Gabriel, and Rio Hondo Rivers. These sediments consist of tongues and lenses of sand and gravel interbedded with silt and clay and are at least 600 m thick [17]. The wells tap a shallow and/or deep aquifer (deepest wells exceed 300 m).

The principal water body is unconfined only within the area which extends a few km south of the Whittier Narrows and which includes the San Gabriel and Rio Hondo Spreading Grounds [18], See Figure 2. One of the main features of the principal water body is the highly permeable Gaspur Zone about 25 m thick, which lies 12 to 25 m below the ground level in the study area. The zone is up to seven km wide and extends from the Whittier Narrows south to the ocean. The spreading ground is the major point of recharge.

The San Gabriel Spreading Grounds, Figures 1-3, are 0.51 km^2 (0.35 km^2 wetted), the intake is 5.7 m^3/s, storage $6.4X10^5$ m^3, and percolation estimated at 2.2 m^3/s. The Rio Hondo Spreading Grounds are 2.3 km^2 (1.8 km^2 wetted), 25.5 m^3/s intake, $3.0X10^6$ m^3 stored, 13.7 m^3/s percolation. In general, little recharge is expected from direct rainfall. The 90 year normal (October 1 to September 30) is 380 mm, but for the years 1959-60, 60-61, 61-62, and 62-63 the rainfall was 59%, 41%, 130%, and 70% of normal, respectively. All the wells have been showing generally declining water levels since the 1930s, and although 1961-62 recharge produced a slight rise, 1962-63 levels were a record low [19,20].

In 1961, the resumption of nuclear testing resulted in much higher tritium levels in rainfall and rivers; consequently, there was a large pulse of tritium introduced in the imported (Colorado River) recharge water and local rain, Figure 4. The estimates of the average tritium concentration were made on the basis of measurements of rain and surface waters over side areas during the last ten years [3,12,13]. It is clear from this curve that the quick upturn due to the resumption of nuclear weapons testing in September 1961, together with the heavier recharge operations using the Colorado River water, which is characteristically higher in tritium than the California rain and river waters, afforded a unique opportunity to test the usefulness of the tritium hydrology method on a large scale and under actual field conditions. The data in Figure 3 give the tritium measurements before September 1961, and later, as the recharge occurred. The tritium increases can be seen mostly near the spreading grounds, and in general a good agreement with the groundwater contours and Free/Pressure level boundary is observed, Figures 3 and 5. The rain in this period was about 100 TU, some of which was directly added, depending upon run-off and the fraction immediately reevaporated. The wells distant from the spreading areas seem to indicate that this may

Figure 1

Downey Plain study area near Los Angeles, from plate 6, USGS Water Supply Paper 1471. Water level contours are for April 1941. Water level contours in feet (0.36 meters = 1 foot). Shaded area is water bearing zone.

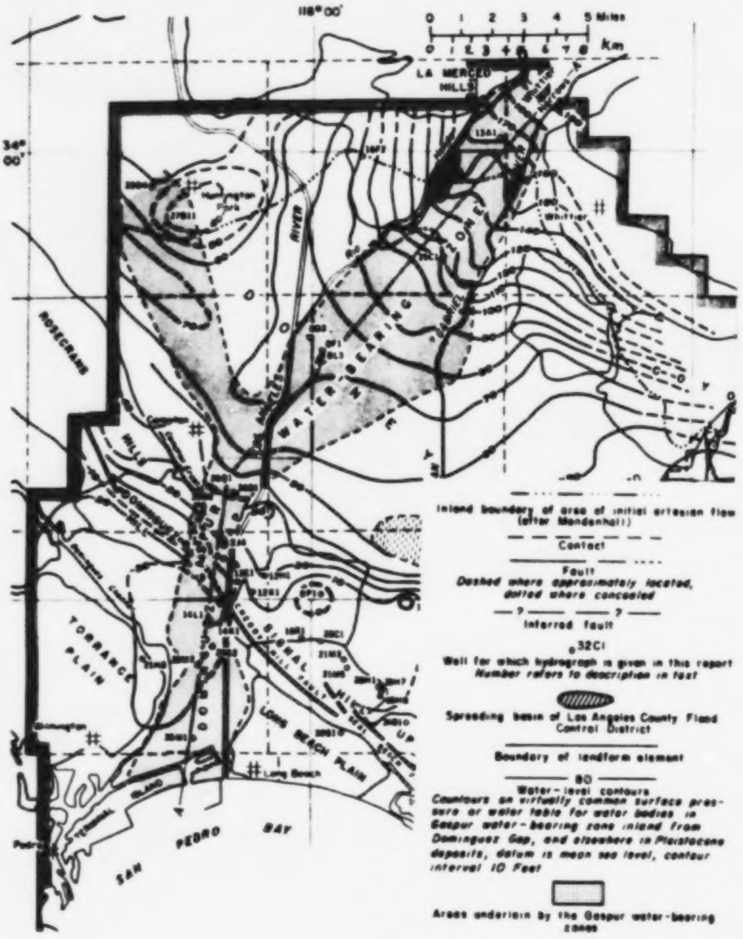

118°00'
0 1 2 3 4 5 Miles
0 1 2 3 4 5 6 7 8 Km
LA MERCED HILLS
34° 00'
RIVER
ROSECRANS
HILLS
WATER-BEARING ZONE
Whittier
TORRANCE PLAIN
LONG BEACH PLAIN
Long Beach
SAN PEDRO BAY
32C1
Inland boundary of area of initial artesian flow (after Mendenhall)
Contact
Fault
Dashed where approximately located, dotted where concealed
Inferred fault
Well for which hydrograph is given in this report
Number refers to description in text
Spreading basin of Los Angeles County Flood Control District
Boundary of landform element
80
Water-level contours
Contours on virtually common surface pressure or water table for water bodies in Gaspur water-bearing zone inland from Dominguez Gap, and elsewhere in Pleistocene deposits, datum is mean sea level, contour interval 10 Feet
Areas underlain by the Gaspur water-bearing zones

Figure 2

Cross section A^1A of study area (see Figure 1) showing area of free recharge and Gaspur Zone. From USGS Water Supply Paper 1471. Altitude in feet (0.36 m = 1 foot).

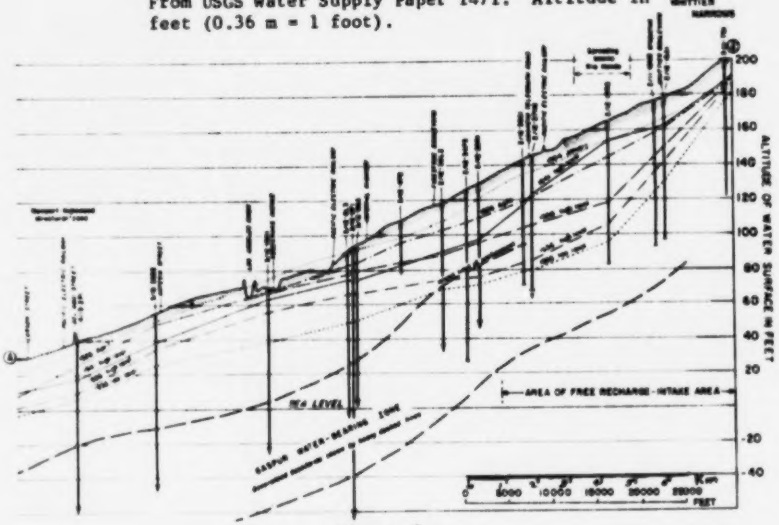

WHITTIER NARROWS
200
180
160
140
120
100
80
60
40
20
SEA LEVEL
-20
-40
ALTITUDE OF WATER SURFACE IN FEET
AREA OF FREE RECHARGE-INTAKE AREA
GASPUR WATER-BEARING ZONE
0
5000
10000
15000
20000
25000
FEET

Figure 3

Tritium measurements in study area. Perforation level of wells is in feet below ground (1 foot = 0.36 meter). Note area of major Tritium addition (heavy solid line).

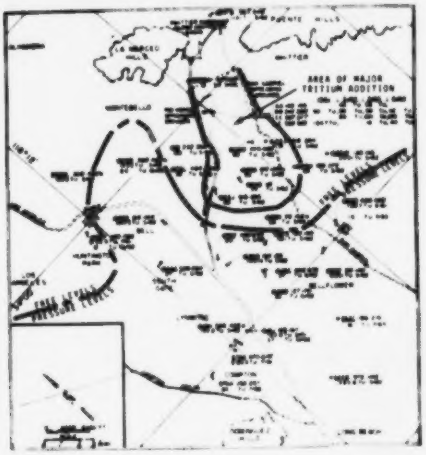


Figure 4

Tritium addition from all sources to the spreading grounds. 1 acre-foot = 1.6×10^3 m^3.

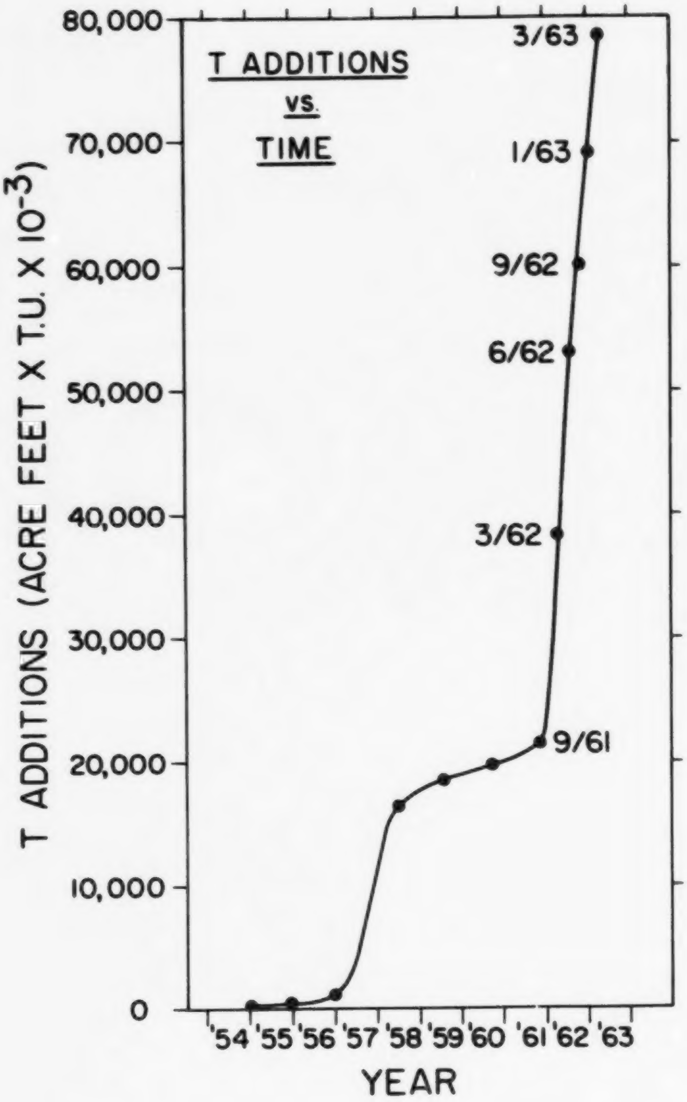
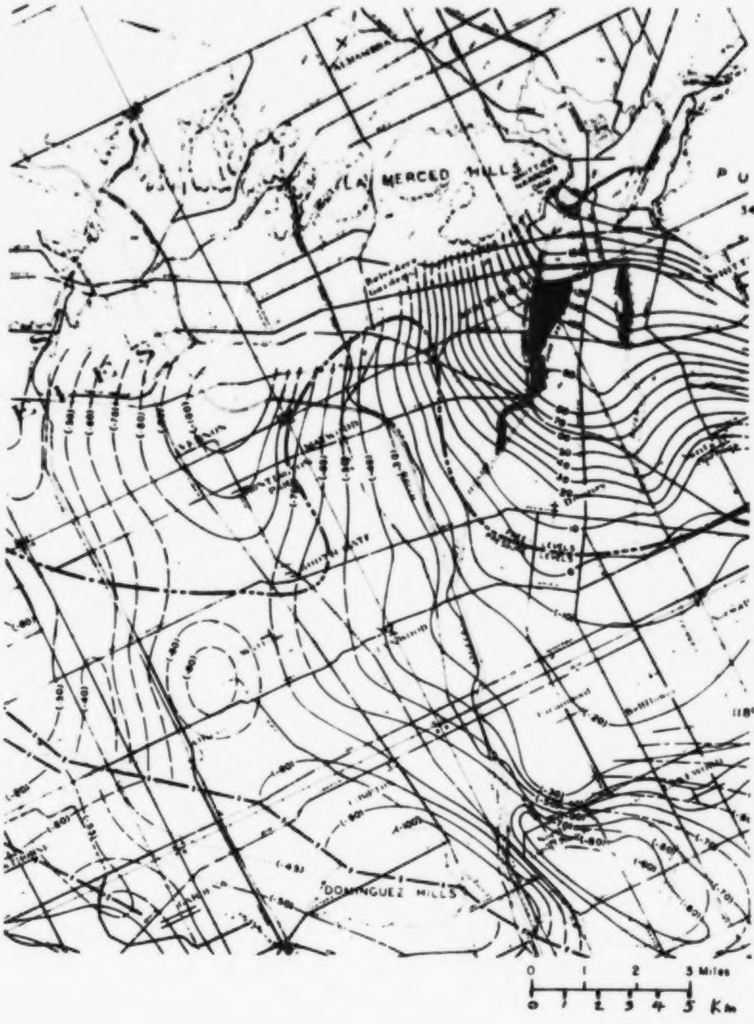


Figure 5

Ground water contours. Map from LA County Flood Control District, Nov. 1962 in Biennial Report, July 1964. Contours in feet (1 ft. = 0.36 m). Note change from 1941 (Figure 1). Area is same as in Figure 3.



amount to as much as 10 to 20 TU, so we may be justified in taking higher levels as evidence of recharge water.

The data show the greatest tritium in the areas where the most recharge is occurring. This area is between and adjacent to the Rio Hondo and San Gabriel Rivers and northeast of the Free-Pressure level boundary, Figure 3. Most of the wells in this area gave 35 TU or more by March 1962, the only exception being the deepest well at the spreading ground (1600Z) which did show 44 TU by June 1963. Outside of this recharge area, only one well shows 35 TU or more (1548B, 35 TU). Actually only four wells outside the recharge area show more than 25 TU: 1504A, 27TU; 1548C, 27TU; 1548B, 35TU; and 1588N, 26TU. Well 1588N may be explained easily since it is a shallow well next to the unlined San Gabriel River. The other three wells cannot be accounted for.

The tritium recharge area correlates with the "Gaspur Zone" boundary between the rivers (Figure 1) and with the "Area of free recharge - intake area" (Figure 2) adjacent to the Whittier Narrows. The results seem to imply that recharge correlates with proximity to the spreading grounds and the upper permeable boundary of the Gaspur Zone. These results imply that horizontal movement from the spreading grounds is more important than vertical percolation. These results are in agreement with the Biennial Report [20] where it is estimated that "the replacement of natural waters with Colorado River water from 1952 to 1962 covers an area of 43 km^2 (a result of importation of 1.05 X 10^9 m^3 of Colorado River water introduced to the underground)."

Sierra-Owens Valley Area

A cooperative program with the Los Angeles Department of Water and Power was undertaken in 1965 to explore the uses of tritium in predicting run-off and recharge in the Eastern Sierra Nevada Mountains-Owens Valley in east-central California, Figure 6. This area is a major source of water for the city of Los Angeles.

There are actually several basins east of the Sierras, including Mono, Long Valley and Owens Lake. These basins are filled with up to 450 m of alluvial material of volcanic and clastic rock (mixtures of gravel, clay and sand). Direct rainfall to the valley floor is small (less than 250 mm) and most of the water is due to run-off, stream flow and underground flow from the melting snow on the eastern slope of the Sierra Nevada Mountains (rainfall equivalent to 1300 mm or more). Much of the underground flow is by percolation through the glacial materials and alluvial fans on the slopes of the Sierra Nevadas [21,22].

The 1962-63 and 1963-64 winter tritium levels in precipitation were higher than in all other years due to nuclear tests in 1961 and 1962. The tritium content of the precipitation in the winter of 1964-65 was less than the previous year, and by 1966-67 the winter precipitation was decreased further [4]. By measuring the tritium content of streams before (April-May) and during (June-July) the maximum spring runoff, we have made estimates of the contribution to the stream flow by the snowmelt of the previous winter (Figure 7 and Table 1).

The tritium changes are correlated qualitatively: a material

Figure 6

Eastern Sierras and Mono-Owens Basins. Sampling points indicated. (From LA Dept, of Water and Power). See inset map of California for location.

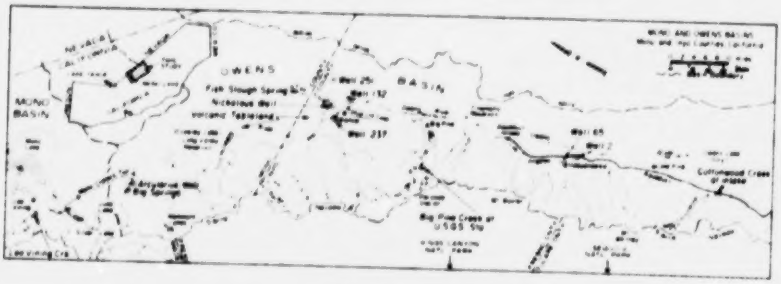


Figure 7

Flow of creeks; arrows indicate date of sampling. Data from LA Department of Water and Power. (1 cubic ft. per second = 28.3 l/sec) Arrows indicate time of sampling for tritium measurement.

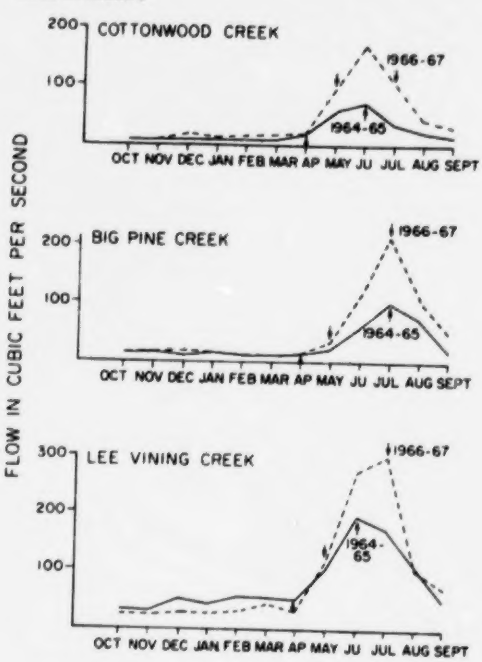


Table 1

Tritium and flow in Sierra creeks

	Cottonwood Creek (at intake)	Big Pine Creek (USGS Station)	Lee Vining Creek (2 1/2 mi. above Ranger Station)
TU 4-65	480	290	535
Flow (cfs)* 4-65	16.6 (9.7)	12.8 (7.5)	50.6 (30)
TU 6-65	-	450	610
Flow 6-65	68.2 (40)	53.8 (31.6)	192.3 (113)
TU 5-67	200	160	200
Flow 5-67	90.3 (53)	35.2 (20.7)	116.3 (68.3)
TU 7-67	240	140	155
Flow 7-67	105.2 (62)	214.5 (126)	297.0 (175)

*cubic feet/sec = 1.7 cubic meters/min

Numbers in parentheses are flow in m^3/min

balance requires that

$$(T \cdot F)_{\text{base}} + (T \cdot F)_{\text{added water}} = (T \cdot F)_{\text{max}}$$

This equation may be solved for the tritium content of the added water

$$(T)_{\text{added water}} = \frac{(T \cdot F)_{\text{max}} - (T \cdot F)_{\text{base}}}{(F)_{\text{added water}}}$$

where T is the tritium content, F is the flow, max is the time of maximum flow (June 1965, July 1967) and base is base flow (April 1965, May 1967). The calculated tritium content (T) of the added water is given in Table 2. These values may be compared to observed tritium precipitation: the estimated TU in rain and snow in the Sierras in 1964-65 was approximately 600 and that observed in 1966-67 was approximately 130 [4,12]. These data indicate that Big Pine and Lee Vining Creeks show direct addition of last winter's precipitation in the spring run-off. Cottonwood Creek tritium data for June 1965 were lost, and the 1967 samples were taken too late to give the pre- and post-melt samples.

The creeks show a tritium increase in 1965, but a decrease in 1967. This is due to the melt of the snow of the previous winter. In 1964-65, the rain and snow were higher in tritium than the creeks; by 1966-67 the creek groundwater was still higher in tritium than the rain and snow. Cottonwood Creek doesn't follow this pattern, due to the sampling and analysis problems. Other hydrologic data on the creeks is given in Table 3.

The wells, Table 4, show varying degrees of recharge and infiltration of surface water. Well 237, which shows high tritium content, has been pumped only intermittently. Since it is sealed, direct infiltration is unlikely, but it is located adjacent to the Bishop Creek Canal and influent seepage from this source probably accounts for the tritium observed.

Table 2

Calculated tritium in Sierra creeks

	Cottonwood Creek	Big Pine Creek	Lee Vining Creek
Calculated TU for water added in 6-65 melt	---	500	637
" 7-67	(437)*	136	126

* See Text

Table 3

Hydrologic data for Sierra-Owens Valley study area

	1964-65	1966-67
Cottonwood Area		
(1) Rainfall at 3200 m elevation	417	544
(2) Water Content at 3300 m elevation	228	635
(3) Flow	$24 \cdot 10^6$	$39 \cdot 10^6$
(4) "Melt"	71%	81%
Big Pine Area		
(1) Rainfall at 2500 m elevation	272	386
(2) Water Content at 3000 m elevation	254	865
(3) Flow	$34.5 \cdot 10^6$	$61.4 \cdot 10^6$
(4) "Melt"	54%	85%
Lee Vining Area		
(1) Rainfall at 2100 m elevation	277	676
(2) Water Content at 2900 m elevation	735	940
(3) Flow	$90 \cdot 10^6$	$102 \cdot 10^6$
(4) "Melt"	41%	66%

(1) At the station nearest the sample collection point, mm, July to June.
(2) Average water content, mm, from snow survey measurements (for 2 or 3 closest stations).
(3) Total flow, m^3 (see Figure 7).
(4) % of total flow above base flow during the spring snow melt period.

Data courtesy of L.A. Department of Water and Power.

Table 4

Tritium data for Sierra-Owens Valley study area

Location	Date	TU	Notes
Lake Crowley	7-63	420	
Silver Lake	11-66	300	
	11-67	150	
Mono Lake	7-63	145	
	6-65	205	
	7-67	190	
	11-67	140	South shore
Hot Spring feeding lake at south shore	11-67	<28	
Big Springs	6-65	70	
	5-67	65	
	7-67	61	
Well 237	5-67	210	16°C, no flow
Fish Slough Spring #1	8-67	100	1.27 cfs*
Well 65	5-67	79,81†	Independence Region, no flow
Arcularius Well	8-67	41,38†	
Well 132	5-67	34	21°C near Bishop, flow 1.3 cfs; Artesian
Nickolous Well	8-67	<18	
Well 251	4-65	<8.5	North of Laws, 2.9 cfs
	5-67	<15	No pumping
Well 2	4-65	<7.0	Flowing, Independence Region, 0.32 cfs
	5-67	<22	Flowing, Independence Region, 0.37 cfs

*cubit feet/sec = 1.7 cubic meters/min.

†duplicate count on enriched sample.

Well 65 was last pumped in 1964, and our sample probably represents water from an upper free aquifer zone, which would contain surface infiltration. Well 2, in the same area, was flowing and probably more nearly represents the tritium content for the lower artesian zones of the wells in the area.

Arcularius Well is adjacent to the Owens River and both the Owens River and the Big Springs source could provide water with tritium.

The interpretation of all the results is impossible without more tritium analyses, but the general picture of low tritium content in the lower aquifer, and surface infiltration from river and canals in the upper aquifer seems to be established. The low tritium content of Big Springs and Fish Slough Springs indicates that the average age of these waters is more than ten years. The source of these springs is thought to be alluvial material southwest of Big Springs and west of Fish Slough.

Since the entire area from Owens to Mono Lakes is shattered by a complex pattern of faults, [21], it is impossible to generalize the groundwater flow pattern because it is probably governed by local faulting and interbedded permeable and impermeable layers.

ACKNOWLEDGMENTS

We wish to acknowledge the cooperation of Mr. Harry Kues and Mr. Charles Green II of the Los Angeles County Flood Control District. Also, we wish to thank personnel of the Los Angeles Department of Water and Power for the collection of samples, and especially Mr. Eldridge Lowry and the late Mr. Charles Dommes, who initiated this portion of the study and Mr. Byron Weinstein and Mr. Paul Lane who provided the hydrologic data and supplementary information.

This research was supported in part by the Chemical Directorate of the United States Air Force Office of Scientific Research, the (University of) California Water Resources Center, and the University of California Research Committee.

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CHAPTER TEN
Tritium Health Physics

RADIOBIOASSAY AT THE KARLSRUHE NUCLEAR RESEARCH CENTER DURING THE YEARS 1967 - 1970

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Radiobioassay serves the purpose of controlling the observance of radiation protection measures. As soon as the body burden surpasses specific limits, working conditions must be changed to avoid further incorporations or at least to reduce them to an acceptable level.

For tritium, in particular for tritiated water generated in heavy water moderated reactors, radiation protection measures consist of keeping the concentration of tritium in the respiratory air below the maximum permissible value of 2 pCi/ml. This is done with adequate ventilation of the work area. In the absence of this precaution, the worker must be protected from exposure by respirators or protective suits. Measurement of the concentration of tritium in respiratory air by appropriate monitors is necessary, since this concentration determines the number of hours the worker may spend in the work area. The hours should be arranged to safely avoid exposures exceeding the maximum permissible continuous body burden.

Using man as a biological indicator, i.e., adopting health physics measures after the events, is a method which should never be adopted. Hence, postexposure surveillance through urine analyses should be used only to confirm the success of the measures and for additional control.

In this report, an account will be given of the experience gathered at the Karlsruhe Nuclear Research Center with respect to Radiobioassay.

Determination of the Minimum Activity Detectable by the Analytical Method Used

The method of detection to be used in routine analyses should be as simple and timesaving as possible. Expensive preparation procedures should be avoided. According to the Recommendations of the International Commisssion on Radiological Protection (ICRP), Publication 10, at least 1/20 of the maximum permissible activity should be detectable, which is about 1.2 μCi of tritium per liter of urine for 500 μCi of tritiated water in the body.

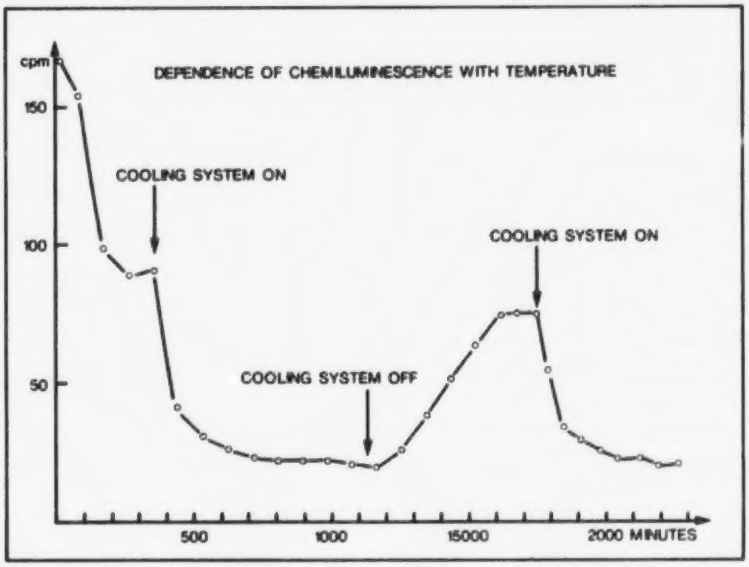
Using a liquid scintillation spectrometer and a scintillator consisting of 4 g 2,5-diphenyloxazole (PPO), 0.05 g 2,2'-p-phenylenbis (4-methyl-5-phenyloxazol) (dimethyl POPOP), 120 g naphthalene for scintillator purposes, and one liter dioxane p.a. [1], the smallest detectable quantity can easily be reduced to 1 nCi/ml. Extended periods of measurement and larger sample volumes allow even low-level detection of tritium in urine with detection limits of 0.2 nCi/l [1]. It was questioned whether this sensitivity of measurement was reasonable in radiation protection for radiobioassay of persons working at the Nuclear Research Center. In this range of measurement, disturbing effects may occur, as was learned from experience.

In the urine of some unexposed persons, count rates were measured which could not be attributed to tritium. In the direct measurement of urine, chemiluminescence is generated; this was considered as the major source of the erroneous counts. As a result of pathologic changes of metabolism, metabolites seem to be excreted in the urine of some persons; these react chemically either with each other or with the scintillation agents, causing chemiluminescence which is measured at room temperature.

The presence of chemiluminescence in urine samples was confirmed by experiment. The count rate of a freshly prepared urine sample, as represented in Figure 1, is characterized by a strong decrease with time. After 400 minutes, this count rate has approached a limit which is characteristic for a specific temperature. When the sample is cooled, the count rate again decreases, approaching a value which corresponds to the background count rate. Gradual reheating, even if performed after a longer period of time, again raises the count rate to another limit and subsequent cooling reduces it to background.

Figure 1

Dependence of Chemiluminescence with Temperature



It was not possible to elucidate the cause underlying this effect, since it is not observed with urine samples of all subjects. However, when measurements are carried out at room temperature, this effect falsely indicates tritium activities up to 0.1 μCi of tritium per liter of urine and, consequently, greatly interferes with the measurement. These errors could be reduced by cooling the samples during measurement. However, the appearance of chemiluminescence cannot be precluded even at lower temperatures. Consequently, the samples are measured at a temperature of about + 15°C. In addition, each lot of samples is measured at least three times for about 10 minutes. Count rates corresponding to specific activities below 20 nCi/l are considered with reservation only.

Therefore, the limit of detection was arbitrarily assumed to be 23 nCi of tritium per ml or 1/1000 of the maximum permissible continuous tritium body burden. This limit of detection in radiobioassay can be safely adopted since, according to Bond [2], the initially feared hazard to men by the organically bound form of tritium and the resulting longer action of tritium upon the human organism can be neglected. There is no need in radiobioassay to achieve excessively high sensitivities at considerable expense. In most occupational cases, the body burdens will be higher than 23 nCi/ml.

We use 1 ml of urine and add it directly to the scintillator solution described above. The effect of direct sunlight is avoided to prevent phosphorescence. The samples are measured at least three times, in a cycle, so that a greater time interval exists between these three measurements in which the disturbing effects are allowed to fade away. The duration of the measurement is 10 minutes [1].

In addition to radiobioassay of exposed persons, the surveillance of the unexposed population becomes more and more interesting. Here, urine analyses cannot be performed in the manner described, because it is too insensitive.

We employ the method of liquid scintillation measurement proposed by Lieberman and Moghissi [3] and use Instagel (Packard) as the scintillator solution to which 10 ml of a distilled urine sample can be added. However, we are faced with the difficulty that, meanwhile, there is no more water which is free from tritium. We have adopted the practice of considering pure scintillator solution as the background. The role of chemiluminescence in this low-level range of measurement is still controversial, which means that a final limit of detection cannot yet be stated. Work on this subject is still going on.

Determination of Tritium Equivalent Doses from Urine Analyses

The equivalent dose D for the whole body can be calulated from the tritium concentration A of urine according to equation (1), assuming that tritiated water is uniformly distributed in the body water [1].

$$D\ [\text{mrem/day}] = 0.51 \times A \qquad (1)$$

To obtain the factor 0.51, the value of "effective energy of tritium for total body EF(RBE)n = 0.010 MeV," as stated in reference [4] (corresponding to a Q-value of 1.7), is multiplied by the converting factors

3.7×10^4 = dis/sec per μCi
1.6×10^{-6} = ergs/MeV
8.64×10^4 = sec/day
10^{-3} = kg/g

when A is expressed in μCi/l urine or body water.

If urine samples are analyzed daily for tritium, the annual dose D_a can be calculated, using modified equation (1) for the number of days per year as follows:

$$D_a \text{ [mrem/a]} = 0.51 \times 365 \times A_m \qquad (2)$$

where A_m represents the average value of all urine measurements, expressed in μCi/l urine.

The practice of daily urine measurement, as described for instance by Osborne [5], is not applied everywhere. Mostly, there is a period of several days between the individual measurements, and sometimes only random samples are taken at very long intervals. This implies that in calculating the dose, the elimination of tritiated water from the body and a possible new incorporation at a specific time must be taken into account.

For one single incorporation, the total equivalent dose is calculated from one urine analysis by integration of equation (1) over time

$$D_\infty = 0.51 \int_0^\infty A \times e^{-\lambda_b t}\, dt \qquad (3)$$

This results in

$$D_\infty \text{ [mrem]} = 0.74 \times T_b \times A \qquad (4)$$

where A is the initial tritium concentration in urine, expressed in μCi/l, $\lambda_b = \ln 2/T_b$ (T_b represents the biological half-life in days) and t is the time in days.

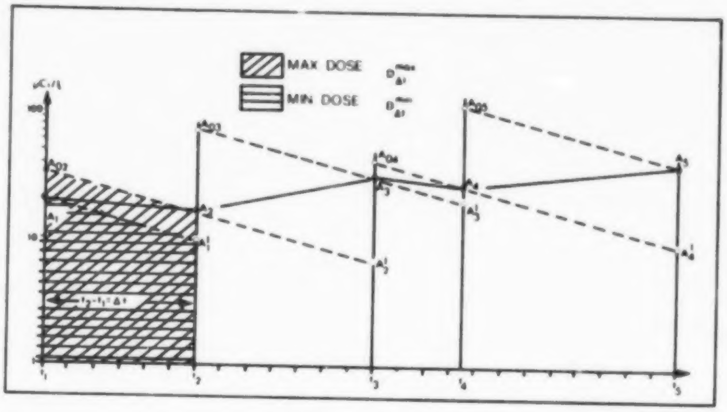
However, in most cases tritium incorporation will not be a unique event but an irregular sequence of individual incorporations. This implies that a dose calculation according to Eq. (4) cannot be performed on the basis of individual analyses and that regular urine analyses must be carried out. Considerations were concerned with the frequency required for such urine analyses to allow adequate radiobioassay and, hence, dose estimate.

If a urine analysis has revealed a certain activity of tritium, this activity will decrease with time according to the biological half-life of tritium. If, upon repetition of the analysis A_1, see Figure 2, a tritium activity is found which is higher (A_2) than the expected value A_1', a pessimistic assumption will lead to the conclusion that an incorporation took place the day after the last urine analysis. It should be large enough (A_{02}) to decrease to the value A_2 found in the second analysis in the interval $t_2 - t_1 = \Delta t$ between the two analyses.

According to the optimistic assumption, the incorporation would have taken place the day preceding the last sampling. This may,

Figure 2

Example for calculation of equivalent doses
from tritium concentration in urine



therefore, result in two different dose values, when dose has to be calculated for the period between t_1 and t_2. With this optimistic assumption, there will be a minimum dose because the second control-measurement by urine analysis will not be included in the calculation. This "minimum dose," $D^{min}_{\Delta t}$, is calculated from the value A_1 by modifying equation (3) and taking the time integral between $t_1 = 0$ and $t_2 - t_1 = \Delta t$ only, as described below:

$$D^{min}_{\Delta t} = 0.51 \int_{t=0}^{t=\Delta t} A_1 \, e^{-\lambda_b t} \, dt \qquad (5)$$

$$D^{min}_{\Delta t} = \frac{0.51}{\lambda_b} A_1 \left(1 - e^{-\lambda_b \Delta t}\right) \qquad (6)$$

According to the pessimistic assumption, an incorporation would have taken place the day following the last sampling, which would have resulted in a hypothetic concentration in urine of A_{02}. Thus, from the value A_{02} a maximum dose D^{max}_t can be calculated for the time between t_1 and t_2, where the value A_1 can be left unconsidered. A_{02} can be be calculated from A_2, using the biological half-life $T_b = \ln 2/\lambda_b$, as follows:

$$A_{02} = A_2 \times e^{\lambda_b \Delta t} \tag{7}$$

The maximum dose $D^{max}_{\Delta t}$ is calculated similarly to equation (5), viz.:

$$D^{max}_{\Delta t} = 0.51 \int_{t=0}^{t=\Delta t} A_{02} \times e^{-\lambda_b t} \, dt \tag{8}$$

This results in

$$D^{max}_{\Delta t} = \frac{0.51}{\lambda_b} A_{02} \left(1 - e^{-\lambda_b \Delta t}\right) \tag{9}$$

and finally, using equation (7), in

$$D^{max}_{\Delta t} = \frac{0.51}{\lambda_b} A_2 \left(e^{\lambda_b \Delta t} - 1\right) \tag{10}$$

The dose values from equations (6) and (10) will be expressed in mrem/Δt, if A_1 and A_2 are specified in μCi/l, and λ_b in day^{-1}.

The difference between the maximum and minimum doses are the greater, the higher, the second value, and the longer the interval Δt between the two urine analyses. A criterion must be found which fixes this interval in such a way as to permit sufficiently accurate radiobioassay and dose estimate.

The biological half-life of tritium of about 10 days is a reasonable limit to the interval in which to carry out radiobioassay: for this reason, urine samples are examined on a weekly basis at the Karlsruhe Nuclear Research Center if an exposure must be anticipated as a result of work performed. However, routine surveillance is performed monthly.

Calculation of the body doses according to (6) and (10) requires the calculation of e-functions, which involves more expenditure. For this purpose, a programmable desk calculator is used. To make calculations easier, the terms

$$K_{min} = \frac{0.51}{\lambda_b} \left(1-e^{-\lambda_b \Delta t}\right) \text{ and } K_{max} = \frac{0.51}{\lambda_b} \left(+\lambda_b \Delta t - 1\right)$$

were calculated for $T_b = 10$ days and for Δt from 1 to 60 days and are compiled in Tables 1a and 1b.

The computing program is conceived in such a way that only the tritium concentrations A_1, A_2, A_3, etc., and the constants K_{min} and K_{max} must be introduced. The constants applicable to the respective interval Δt between the individual analyses can be taken from Tables 1a and 1b. The mean value between maximum and minimum dose is considered to be probably the proper dose, when more specific data on the exact time of incorporation are not known.

With the help of this computing program, the necessary dose calculations are readily performed by one person. However, this

Table 1a

Calculation of K_{min} and K_{max}

Δt (days)	$K_{min} = \frac{0.51}{\lambda_b}\left(1 - e^{-\lambda_b \Delta t}\right)$	$K_{max} = \frac{0.51}{\lambda_b}\left(e^{\lambda_b \Delta t} - 1\right)$
1	0.493	0.528
2	0.952	1.094
3	1.381	1.700
4	1.781	2.351
5	2.155	3.048
6	2.503	1.156
7	2.828	4.595
8	3.131	5.453
9	3.415	5.066
10	3.679	7.358
11	3.925	8.414
12	4.155	9.546
13	4.370	10.759
14	4.570	12.059
15	4.756	13.453
16	4.930	14.946
17	5.093	16.548
18	5.245	18.263
19	5.387	20.102
20	5.518	22.072
21	5.641	24.185
22	5.757	26.449
23	5.863	28.875
24	5.963	31.476
25	6.057	34.263
26	6.144	37.251
27	6.225	40.452
28	6.301	43.884
29	6.372	47.561
30	6.438	51.502

Calculated on the basis of ($\lambda_b = \frac{\ln 2}{T_b}$ and T_b = 10 days).

Table 1b

Calculation of K_{min} and K_{max}

Δt (days)	$K_{min} = \frac{0.51}{\lambda_b}\left(1 - e^{-\lambda_b t}\right)$	$K_{max} = \frac{0.51}{\lambda_b}\left(e^{3\lambda_b t} - 1\right)$
31	6.499	55.727
32	6.556	60.255
33	6.610	65.108
34	6.660	70.309
35	6.707	75.883
36	6.751	81.857
37	6.791	88.261
38	6.829	95.123
39	6.865	102.479
40	6.897	110.362
41	6.928	118.811
42	6.957	127.867
43	6.984	137.572
44	7.009	147.974
45	7.032	159.123
46	7.206	171.072
47	7.074	183.878
48	7.094	197.604
49	7.111	212.314
50	7.127	228.081
51	7.143	244.978
52	7.157	263.089
53	7.171	282.500
54	7.183	303.304
55	7.195	325.600
56	7.205	349.498
57	7.216	375.110
58	7.225	402.562
59	7.234	431.982
60	7.242	463.515

Calculated on the basis of ($\lambda_b = \frac{\ln 2}{T_b}$ and $T_b = 10$ days).

relatively expensive calculation is done only in cases where more than 10 percent of the maximum permissible continuous body burden is measured. If the tritium concentration found in the urine is always less than 10 percent of the maximum permissible body burden, the dose is estimated according to Eq. (2). Using this equation, one may safely assume that an excessive incorporation would have been detected.

Results

In the years from 1967 to 1970, three groups of workers were surveyed for tritium. They included the staff employed at the two heavy water moderated reactors FR 2 and MZFR as well as those in the Decontamination Services.

Body equivalent doses were calculated from urine analyses as described in this paper, and their frequency distribution was divided into five logarithmically graded classes. Table 2 and Figure 3 represent the results of the investigations. Percentages refer to the number of persons examined. It appears as a general finding that in no case the maximum permissible continuous burden of tritium in the body was surpassed.

Due to the smaller quantity of tritium handled in the Decontamination Services, the burden of the staff working there is smaller than with personnel in the reactor operating departments. Only a few of them had body burdens of 1 to 10 percent of the maximum permissible level. The majority were practically unexposed.

The reverse tendency is observed with staff working at the two reactors. The fraction of unexposed persons is low; most of the incorporations fall within the range of 1 to 10 percent of the maximum permissible body burden. It is a noticeable fact that the fraction contributed by the group with 10 to 100 percent maximum permissible body burden is higher in MZFR than in FR 2, although the tritium content of MZFR, as shown in Figure 4, had been lower than in FR 2 over the whole period of surveillance.

The higher exposure was caused by early repair and maintenance work. Initially, repairs had been carried out without the use of respirators. This is the reason for the comparatively high incorporation in MZFR in 1967/68. Respirators are now being used for maintenance work, decreasing the level of incorporations in MZFR since 1969. A new increase in incorporation found in 1970 under identical working conditions is primarily due to the increase in tritium content of the heavy water. This requires more rigorous radiation protection measures. In FR 2, recent major repair work on an experimental heavy water loop is the only explanation for a higher incorporation rate. Hence, a comparison with pure power reactors is not possible. In principle, radiation protection measurements were adequate for all work, which was confirmed by regular radiobioassay of the reactor staff. There was no excessive incorporation.

Table 2

Distribution of Equivalent Doses from Tritium Urine Analyses

	Equivalent Doses mrem/a	Distribution in percentage of surveyed persons			
		1967	1968	1969	1970
FR 2	<5	0.0%	15.4%	4.4%	1.1%
	5 - 50	27.5%	20.5%	19.6%	33.8%
	50 - 500	47.5%	41.0%	60.8%	53.5%
	500 - 5000	25.0%	23.1%	15.2%	11.6%
	>5000	0.0%	0.0%	0.0%	0.0%
number of persons		40	39	46	86
MZFR	<5	2.5%	0.7%	5.9%	0.0%
	5 - 50	12.5%	5.6%	24.8%	11.0%
	50 - 500	62.5%	45.8%	55.0%	70.0%
	500 - 5000	22.5%	47.8%	14.3%	19.0%
	>5000	0.0%	0.0%	0.0%	0.0%
number of persons		80	144	238	109
Decont. facility	<5	72.0%	54.3%	70.0%	82.8%
	5 - 50	21.3%	35.4%	27.1%	14.3%
	50 - 500	6.6%	10.3%	2.9%	2.9%
	500 - 5000	0.0%	0.0%	0.0%	0.0%
	>5000	0.0%	0.0%	0.0%	0.0%
number of persons		61	48	60	70

Figure 3

Distribution of equivalent doses to man from tritium-urine-analyses during 1967-1970 (in percentage of surveyed persons)

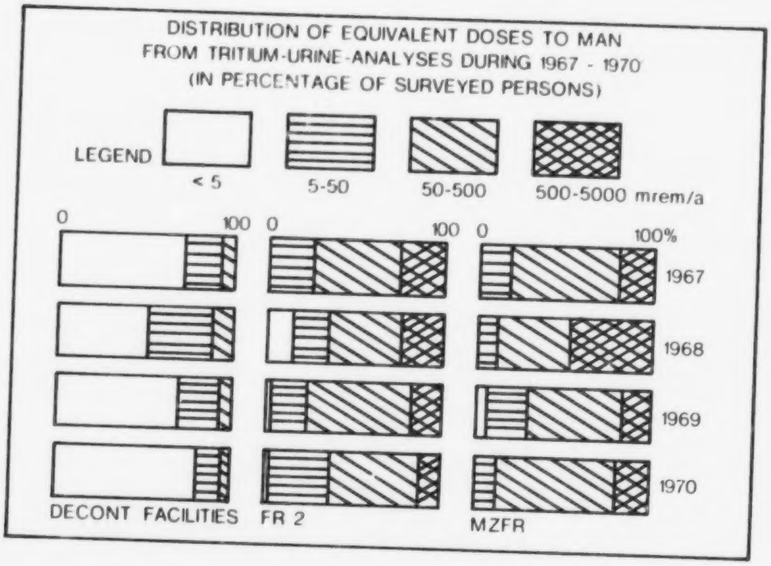
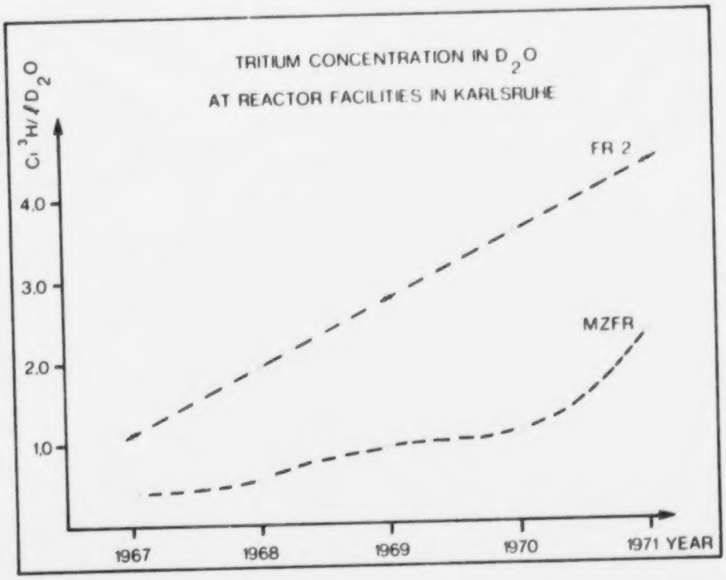


Figure 4

Tritium concentration in D_2O at reactor facilities in Karlsruhe



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PRACTICAL CONTROL OF TRITIATED WATER VAPOR BY SILICA GEL AND OF TRITIUM GAS BY THE USE OF A PURGING DEVICE

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Our need for tritiated water has steadily increased over the past few years. To meet this demand and also to minimize the chance of contamination, a method of handling had to be developed that would meet environmental limitations and yet be flexible enough to accommodate the different types of operations involved.

Tritiated water, with a specific activity of approximately 40 Ci/ml, is shipped to us from the supplier, in glass ampoules containing 20 ml quantities. For reasons of safety and convenience, we subdivide these large volumes into smaller working quantities, usually 2 ml, to meet requirements that vary anywhere from a few lambda upward. This transfer can best be done using regular pipetting techniques in a glove box; vacuum transfer would make accuracy difficult to maintain. However, filters in the effluent air lines of these glove boxes were ineffective in trapping tritium gas and tritiated water vapor released during the operations.

To rectify these insufficiencies, we designed the "degassing unit" that removes and traps the tritium gas released when a storage ampoule of tritiated water is opened. Further, knowing that dehumidification of air streams by silica gel or other desiccants is a common practice [1], we developed a filter using a silica gel that removes the tritiated water vapor from the effluent air of a glove box.

Silica Gel Adsorption

The experimental setup used for evaluating the relative effectiveness of different configurations of silica gel adsorption columns was designed as follows: With flow rate ranging from 6 to 10 l/min created by the pump in the tritium sniffer, the air is drawn from the tritiated water vapor source in the glove box, through an ionization chamber to which a strip-chart recorder is attached, through the test column of silica gel, through the tritium sniffer to which another recorder is attached, and then exhausted. No attempt was made to control the temperature or humidity of the room air.

A test pulse of tritiated water vapor (10 ci/m^3 for 40 min) was released in the glove box and drawn through the system to load the adsorption column filled with either oven-dried silica gel, or silica gel in equilibrium with the ambient humidity. Other desiccants tested included activated alumina and $CaSO_4$. The characteristics of adsorption columns filled with Davisson Grade 40 silica gel (6 to 12 mesh) were further examined by evaluating the effect of various column dimensions under similar air flow rates.

As with other dynamic adsorption systems, this process can be considered diffusional. The equations suggested by Madey et al. [2,3] can be applied to characterize the operation and allow the prediction of behavior.

The basic equation for transmission of an adsorbate through a column is stated by Lee and Madey [3]. For a column of length ℓ and superficial flow velocity (u), the transmission (T) as a function of time (t) is given by

$$2T = \exp\,(1/\Delta)\ \mathrm{erfc}\ S_+ + \mathrm{erfc}\ S_-$$

with

$$2S_\pm = (1/\Delta)^{1/2}[(B\ell/ut)^{1/2} \pm (ut/B\ell)^{1/2}]$$

and

$$\mathrm{erfc}\ S = 1 - (2/\sqrt{\pi})\int_0^S \exp(-x^2)dx.$$

Δ is a dimensionless dispersion number; and B, the "effective adsorptivity," is defined as equal to ρ dK/dv+f, where ρ is the bulk density and f the void fraction of the adsorbent, and dK/dv is the rate of change of the isothermal volume adsorption capacity with respect to the mol fraction of the adsorbate. If the total volume of the adsorbent in the column is V, the product BV can be thought of as the total adsorption capacity of the column. In the case of a short pulse of adsorbate, the maximum concentration in the column effluent will occur after the volume of gas passed through approximately equals the total adsorption capacity. For small values of Δ the agreement is quite close. With larger values of Δ, the maximum will occur sooner than predicted.

Thus, two parameters are sufficient to characterize an adsorption column: a dispersion number and the total adsorption capacity. The adsorption capacity is a property of the adsorbent, but the dispersion number depends on parameters such as column dimensions, flow rates, and particle size.

Figure 1 compares typical breakthrough curves for oven-dried silica gel and silica gel already in equilibrium with the ambient humidity. In either case, no significant amount of tritium shows up in the effluent for more than 25 h.

A few log-log plots are shown in Figure 2, covering a range of column sizes, shapes, and flow rates; all curves have been normalized to an input pulse totaling 1 Ci. Even with poorly designed columns, more than 1000 column volumes can pass through the adsorbent before breakthrough. Reasonable effective designs are good for about 10,000 column volumes. Using the Davisson silica gel, we found an effective adsorptivity of about 35,000. Practical experience has shown that other manufacturer's silica gel of equivalent quality is also satisfactory. The dispersion number varied with different columns. For a column of convenient size, in which the diameter and length are about equal, and a superficial velocity of 10 cm/sec, we obtained a dispersion number of about 0.08. Longer columns and lower velocity tend to decrease the dispersion, up to a point. The sharpest breakthrough curves (lowest dispersion numbers) were obtained with a column 23 cm long and 7 cm in diameter, and a superficial velocity of 4.3 cm/sec. This gave a dispersion number of 0.02.

Figure 1

Comparison of the breakthrough curves for oven-dried silica gel ( ) and silica gel in equilibrium with the ambient humidity (+).

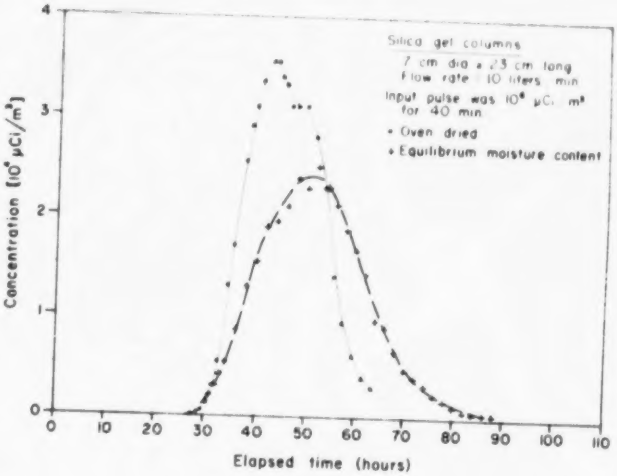
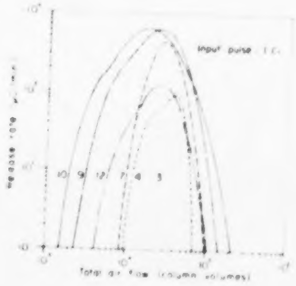


Figure 2

Log-log plots covering results from a range of column sizes, shapes, and velocities.



Column characteristics

Run no	Dia (cm)	Lgth (cm)	Vol (cm³)	Flow rate (ℓ/min)	Superficial velocity (cm/min)
5	7.0	23	885	10.0	261
6	8.9	58	236	6.7	108
7	4.4	5.1	79	9.5	614
9	1.6	20	40.2	9.7	4824
10	4.4	26	395	9.8	633
12	8.9	58	236	10.0	161

If the log of the concentration is plotted against the log of the time, a symmetrical curve should be obtained. Its shape is a function only of the dispersion number, and its location on the x axis is a function only of the adsorption capactiy. The curves we observed approximated this ideal, but in some cases the shape tended toward a double-humped curve. Use of room air may account for the variability and lack of symmetry.

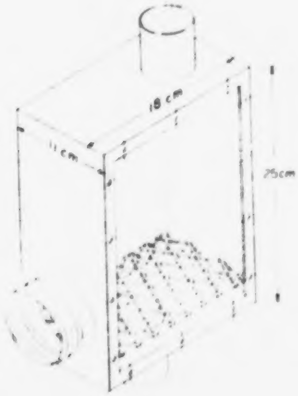
Our studies show that it is not necessary to first dehydrate the silica gel. It can be saturated with water at ordinary room temperature and humidity, and will still remove tritiated water vapor with an effectiveness equivalent to that of the dry desiccant. The reason is that a rapid exchange occurs between water in the adsorbed and vapor phases. The same amount of water is leaving as is entering the column, but it is not the same water. Thus, although tritiated water enters, only normal water leaves, until the column nears saturation with the tritiated water.

Several other desiccants were tried. Activiated alumina and $CaSO_4$ were found to be inferior desiccants for this application. Linde molecular sieve, grade 5A, was about equal to silica gel in effectiveness.

Based on our observations of the adsorption columns, a special silica gel filter was designed, Figure 3, to accommodate the air flow of approximately 280 l/min required by the glove boxes. Calculations showed that for such an air flow 2.25 kg (5 lbs) of silica gel should hold the tritiated water vapor for approximately one hour before it begins to break through. The filters have a volume of approximately five liters, allowing a 2.5-cm plenum between the air intake and the silica gel that is supported by a fine mesh screen resting on an expanded-metal grid. The filters are fitted with a filling spout and a bag pass-out for convenience and economy.

Figure 3

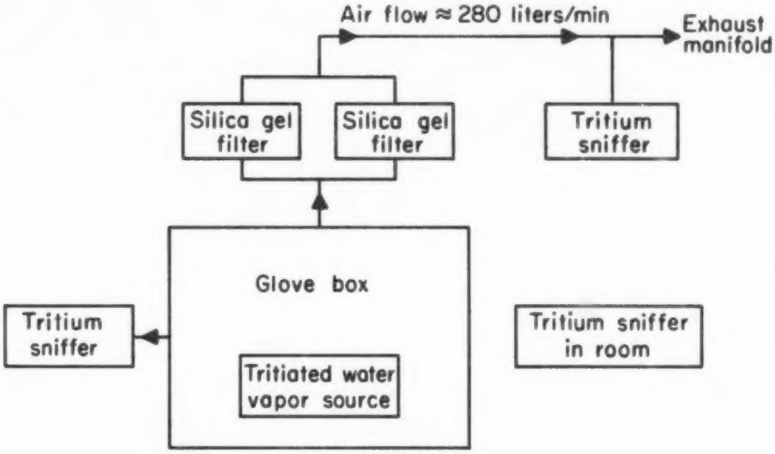
Silica gel filter.



A typical glove box arrangement used in transfer of tritiated water is shown schematically in Figure 4. The two silica gel filters are connected parallel to the box by flexible ducting, but are used alternately, permitting safe and convenient removal of one saturated filter and its replacement while maintaining a steady uninterrupted air flow.

Figure 4

Typical glove box arrangement for transfer of tritiated water.



XBL 725-2855

Tritium Degassing Unit

When the large ampoules of tritiated water are subdivided, the small volumes are placed in storage ampoules which consist of test tubes fitted with a Kovar seal and a metal screw cap. These are stored in groups of ten in brass storage cylinders, Figure 5, that are sealed and held under 38 cm of vacuum.

The tritiated water has a tendency to break down upon standing, to form several gases, one of which is elemental tritium. Since the silica gel does not capture or hold back elemental tritium, the "degassing unit" described below, Figure 6, was designed to eliminate the potential hazard of escaping gas, and is used whenever a storage ampoule is opened.

This degassing unit is placed in the glove box and attached, by Tygon tubing, to an externally located, five-liter vacuum bottle which has been pumped down to a gauge pressure of about -74 cm; the volume of this bottle is optional. The storage ampoule is exposed to vacuum

Figure 3

Arrangement of storage ampoules in storage cylinder. After loading, the top (not in picture) is sealed on and the atmosphere reduced to 38 cm of vacuum.

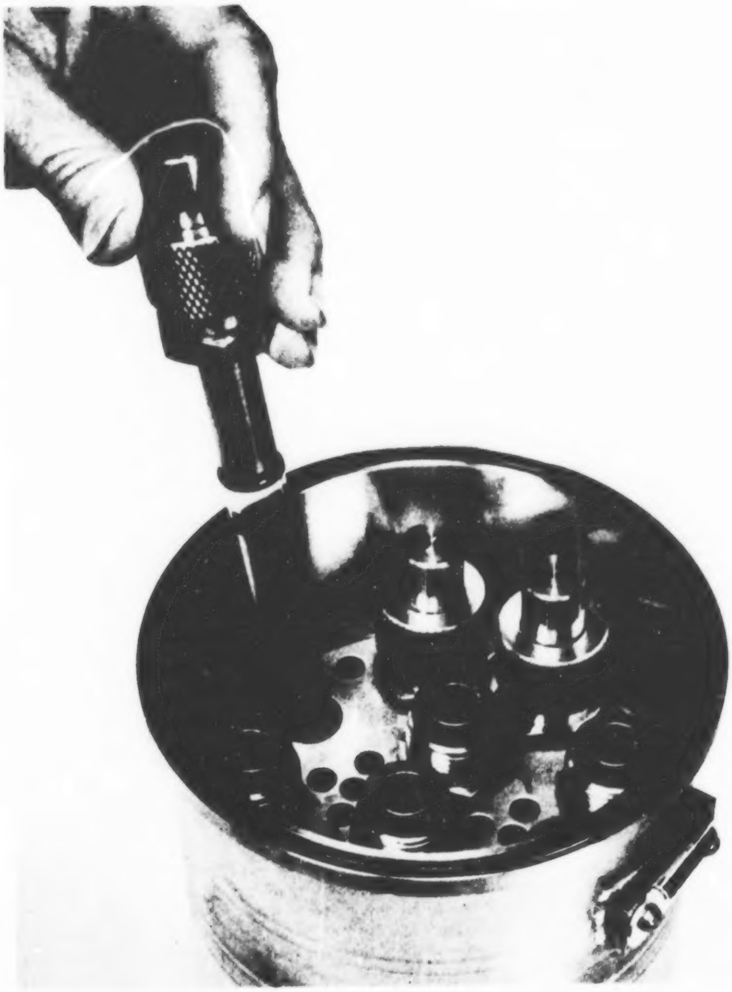


Figure 6

Tritium degassing unit: A. Valve to vacuum bottle; B. Screw cap to ampoule; C. Handle that controls top of ampoule; D. Valve that opens to atmosphere.

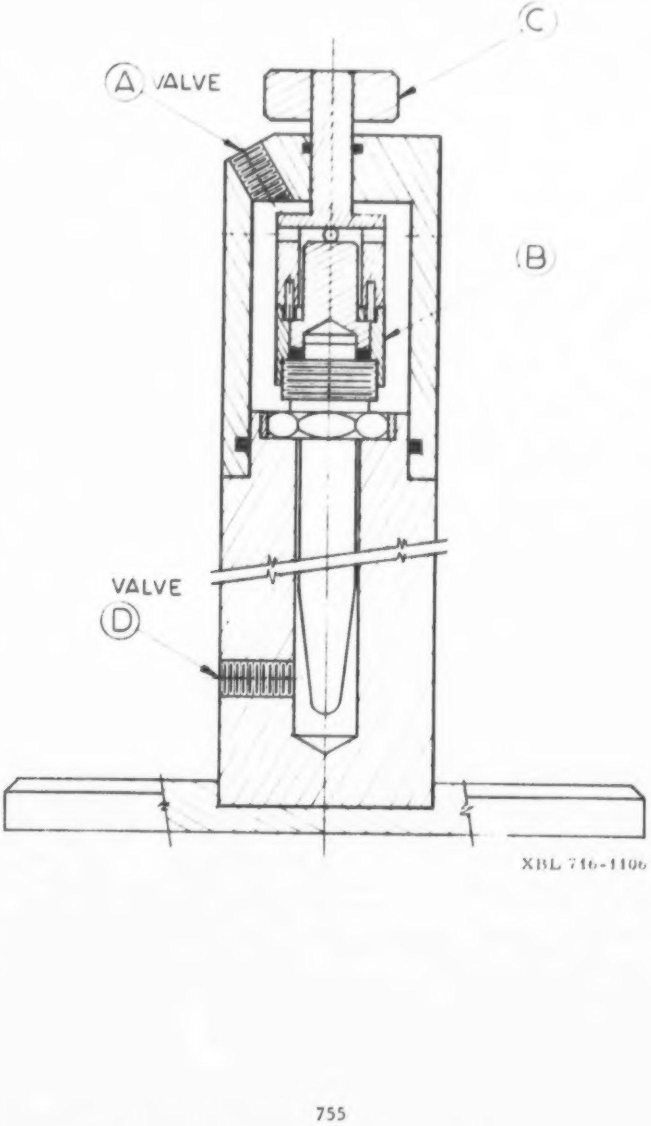


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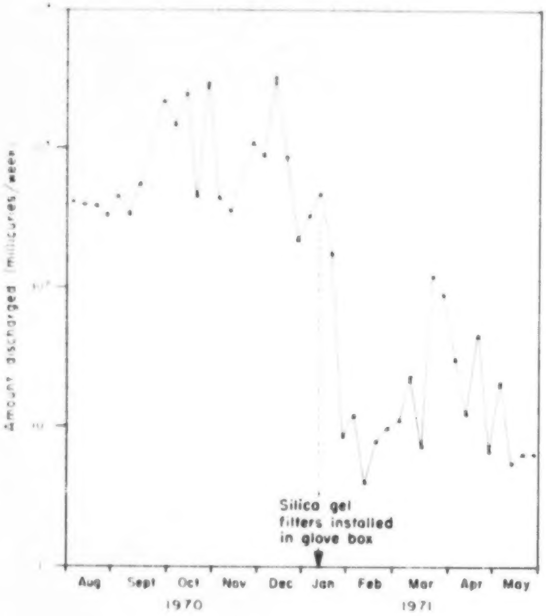
Figure 7

The amount of tritium discharged (mCi/week) before and after the silica gel filters were used.



through. We hope to get a reasonable answer to this in the near future.

ACKNOWLEDGMENTS

We are grateful to Robert M. Latimer, Health Chemistry Department Head, for supporting this project by reinforcing the Health Chemistry policy of minimum release to the environment, and James T. Haley for his many good suggestions and invaluable assistance. We thank Eileen Doyle for her drawings and Carol Legge for her secretarial assistance.

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ASSESSMENT OF RADIOLOGICAL HAZARDS FROM TRITIUM CONTAMINATED MATERIALS - AN APPROACH

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With the development of heavy water moderated and cooled power reactors and an increase in the heavy water reactor installed capacity in the world, the need may arise to ship components from heavy water systems to non-nuclear installations for inspection and/or maintenance. Tritium is produced in large quantities in heavy water by activation, and diffuses into material of the system components. Decontamination of tritium is difficult compared to decontamination of fission and activation products [1]. "Bleed out" is observed because tritium that has diffused into the material diffuses out and, due to its physical properties, becomes airborne. Hence, it is necessary to evolve a simple and fast technique to determine total tritium release from contaminated articles, and to assess the radiological hazards present during shipment, inspection and/or maintenance. Determination of tritium content should also facilitate assessment of radiological hazards, thereby aiding the implementation of proper safety procedures during storage at the plant site and during inspection and/or maintenance by plant personnel. An attempt has been made to estimate total tritium release on the basis of smeared activity from the surface of the experimental specimen under normal ventilation conditions at room temperature. An empirical relationship correlating the smeared activity and the total tritium release is proposed.

Methods

Two plates, each of mild steel, aluminium, polyethylene and rubber, were immersed in heavy water containing 7.5 Ci/l tritium activity for a period of thirty days. Total surface area of each plate was about 419.2 cm^2 (10 cm X 20 cm X 0.32 cm). The mild steel had been conditioned similarly to the mild steel of reactor systems and had a surface coating of black iron oxide. The rubber was gasket materials of primarily natural rubber (polyisoprene). The heavy water was free from fission and activation product activities, except tritium. Exposures were carried out at room temperature (24°C), and heavy water pH was observed to be between 6.5 and 7.

The plates were removed from the heavy water after thirty days of exposure and were wiped to dryness. All were decontaminated by washing for ten minutes in a warm water solution of one percent sodium ethylene diamine tetra-acetic acid (EDTA) and phosphate laboratory detergent (Alconox). After decontamination, the plates were again wiped and dried at room temperature, then separated into two groups for airborne tritium activity and smear activity measurements. Each plate was kept separately in a ventilated polyethylene container, the volume of which was seven liters with a dry air purge flow rate of one l/min measured with a 0 to 2 l/min rotameter.

The water bubbler for trapping airborne tritium consisted of a standard gas washer containing 100 ml of water for all the measurements. Tritium absorption on the components of the experimental setup was observed to be negligible. The procedure followed for each group is summarized in Figure 1. Figure 2 is a schematic diagram of the equipment used for the measurements.

Figure 1

Summary of Experimental Procedure

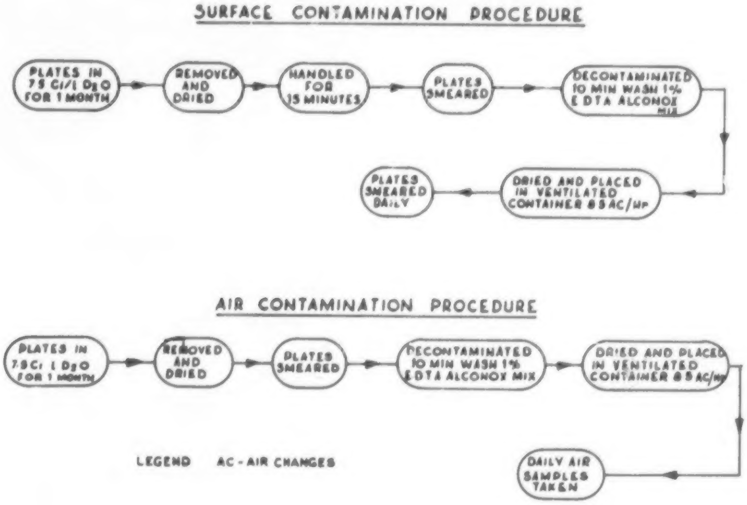
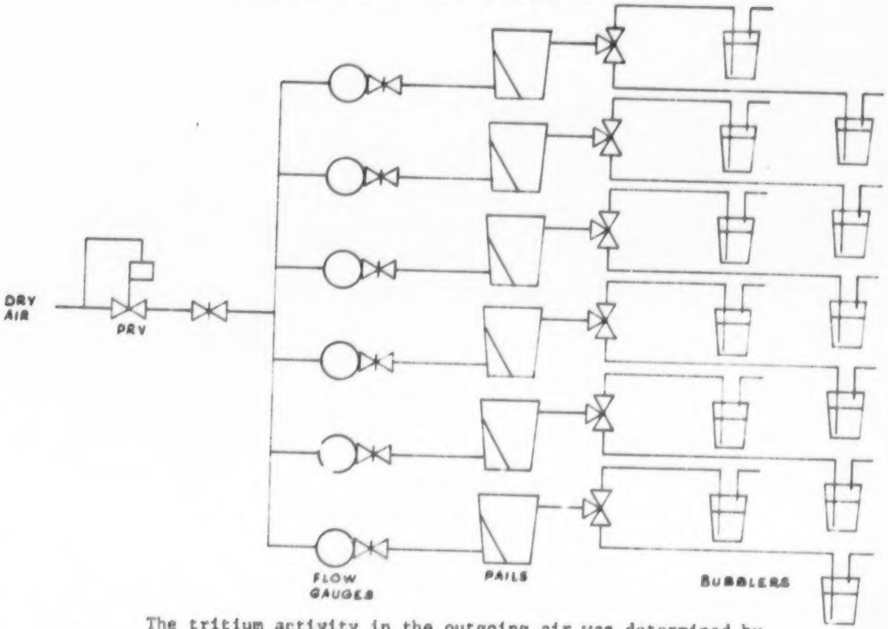

SURFACE CONTAMINATION PROCEDURE
PLATES IN 7.5 Ci/l D_2O FOR 1 MONTH
REMOVED AND DRIED
HANDLED FOR 15 MINUTES
PLATES SMEARED
DECONTAMINATED 10 MIN WASH 1% EDTA ALCONOX MIX
DRIED AND PLACED IN VENTILATED CONTAINER 8.5 AC/HR
PLATES SMEARED DAILY
AIR CONTAMINATION PROCEDURE
PLATES IN 7.5 Ci l D_2O FOR 1 MONTH
REMOVED AND DRIED
PLATES SMEARED
DECONTAMINATED 10 MIN WASH 1% EDTA ALCONOX MIX
DRIED AND PLACED IN VENTILATED CONTAINER 8.5 AC/HR
DAILY AIR SAMPLES TAKEN
LEGEND AC - AIR CHANGES

Figure 2

Equipment for Ventilation Experiment



The tritium activity in the outgoing air was determined by measuring tritium activity in the bubbler water with a liquid scintillation spectrometer, using 1 ml of water sample and 20 ml of liquid scintillator as described below. Samples were collected at various time intervals. In the latter stages of the work, when air contamination levels were low, samples were collected for periods of 24 hours. The measured tritium activity in bubbler water was corrected for evaporation losses of water by a factor $C_t = (V_s + V_e)/2V_s$, where V_s and V_e are the volume of water at the start and end of the sampling time, respectively.

The technique used for smear activity measurement is similar to the normal procedure for the measurement of loose fission product contamination of surfaces. A dry two inch disc of Whatman-42 filter paper was rubbed evenly over both sides of the plate. The paper was then placed in a plastic vial, and 20 ml of liquid scintillator was added. The vial was counted in a Packard Tri-Carb 3000 series liquid scintillation spectrometer in the normal manner. The liquid scintillator solution is that recommended by Butler [2], consisting of 0.05 g 1,4-bis-2-(4-methyl-5-phenyl oxazolyl)-benzene (dimethyl POPOP), 4 g 2,5-diphenyloxazole (PPO) and 120 g of naphthalene dissolved in one liter of p-dioxane. The counting efficiency for these samples ranged from 18.4 to 23.9 percent as determined by an internal standard. An average efficiency of 21 percent was used for all the measurements except for the initial smear of the mild steel plate. The efficiency in this case was determined to be 10 percent.

Results

Variation of airborne activity with time for mild steel, aluminium, rubber and polyethylene is shown in Figures 2 and 3, respectively. The smear activity normalized to the airborne activity follows the same decay pattern for mild steel and aluminium but the agreement is not good for rubber and polyethylene. The activity release for all the materials was estimated over a period of 120 hours, after which the release was found to be small. The total release, including the release after three hours of ventilation until the end of the sampling and the smear activity after three hours of ventilation, is given in Table 1. Release after three hours of ventilation has been computed specifically to coincide with the timing of smear collection.

Figure 3

Variation of Air Activity & Normalised Smear Activity

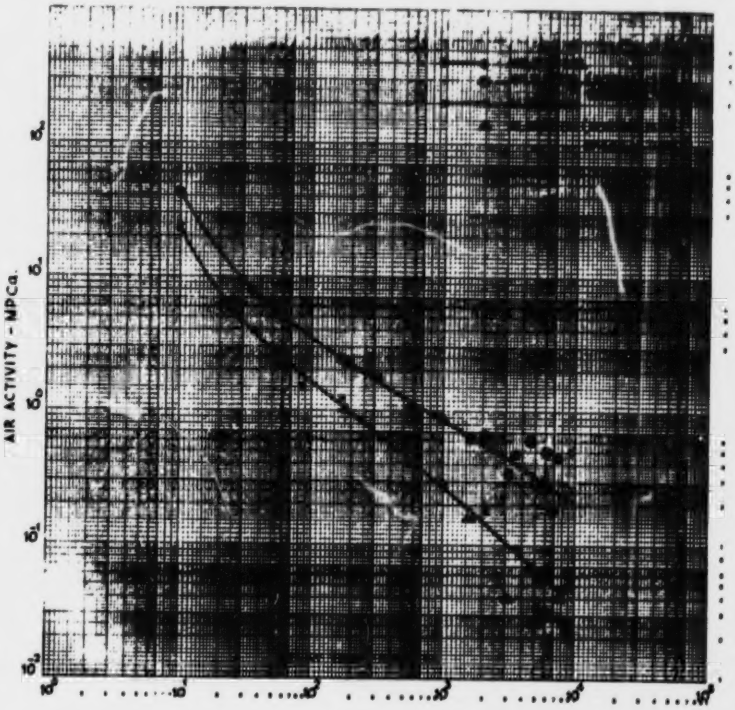


Figure 4

Variation of Air Activity & Normalised Smear Activity

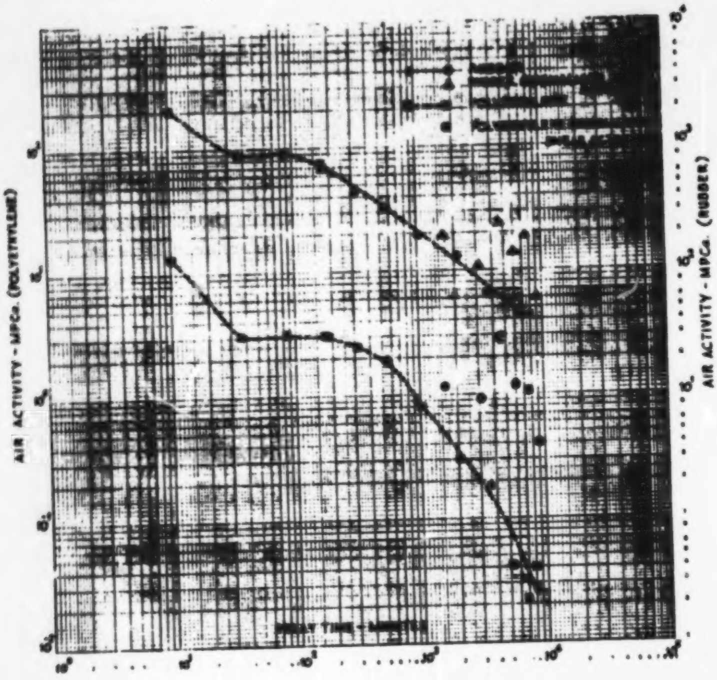


Table 1

Estimated Tritium Release and Smeared Activity

Material	Activity 120 h (µCi)	Release in 117 h (µCi)	Smeared Activity after 3 hours Ventilation (pCi/cm^2)
Mild Steel	23.15	12.47	41.45
Aluminium	8.40	4.93	292.0
Polyethylene	14.90	11.01	8.22
Rubber	4.03 X 10^3	3.87 X 10^3	70.80

The decreased airborne activity in the ventilation stream is plotted in Figures 2 and 3 for the materials investigated. The smear activity is normalized and plotted on the same figures.

Discussion

The airborne and smear activities follow the same decrease pattern for mild steel and aluminium, but for rubber and polyethylene the agreement is not good during the latter part of the measurements. The total activity release p(μCi) from the specimen can be related to smear activity S(pCi/cm^2) at the start of the release time by the following expression:

$$S = kp \qquad (1)$$

where k is the constant of proportionality.

In this relationship, variables S and p are individual functions of diffusion constant, temperature and structure of the material under investigation. Hence, the variation of S and p should follow the same pattern, as has been found in the present work. The most likely varying parameter is temperature; the tritium release will be higher at higher temperatures and, accordingly, the smear activity should also increase.

On the basis of relationship (1), it is possible to propose smear activity levels for unrestricted shipment of articles to non-nuclear installations. For the purpose of indicating an approach to derive the permissible smear activity, we consider 0.12 mCi as the permissible release from the contaminated objects and calculate the smear activity levels for unrestricted release. A sample calculation is given below:

In the case of aluminium:

Smear activity after three hours ventilation
(from Table 1): S = 292 pCi/cm^2

Release from three hours to the end of the experiment
(from Table 1): p = 4.93 μCi
Permissible release: P = 0.12 mCi

From equation (1), the permissible smear
levels are given by:

$$Sp = \frac{P \times S}{p} \qquad (2)$$

Where various terms are as explained above.

Hence, permissible smear level Sp for aluminium is obtained by putting in the values for various terms from above and is given as:

$$Sp = 6.90 \times 10^3 \text{ pCi/cm}^2$$

Table 2 gives the derived permissible smear levels in terms of the surface area of the article. These levels have been worked out for the materials investigated.

Table 2

Derived Permissible Smear Levels
for 0.12 mCi Tritium Release

Material	Smear Activity (pCi/cm^2)
Mild steel	1.01×10^3
Aluminium	6.90×10^3
Polyethylene	8.62×10^1
Rubber	2.17

The results obtained are applicable to a limited number of materials and are derived from an experiment conducted under limited exposure and environmental conditions. However, the principle put forward in establishing permissible smear levels for a possible release of tritium contaminated equipment to a non-nuclear installation is considered practical and feasible.

ACKNOWLEDGMENTS

The author thanks Mr. R. Wilson, Manager, Health Physics Department, Ontario Hydro, Canada for suggesting the problem and continued guidance throughout, and is grateful to the Medical Service Division, Ontario Hydro and Health Physics Division, BARC, for permission to publish this work. The work was done at Douglas Point Generating Station, Ontario, Canada, as part of the training program under the Colombo plan.

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DESORPTION OF TRITIUM FROM TARGET SOURCES

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Thin layers of tritium-impregnated Zr or Ti with suitable metallic backings are being widely used in neutron generators and gas chromatographic studies. The method of preparation and integrity of such targets from a neutron-yield point of view has been reported earlier [1-5]. The data available on the dissociation of tritium from target surfaces and the associated radiological hazards are limited [6].

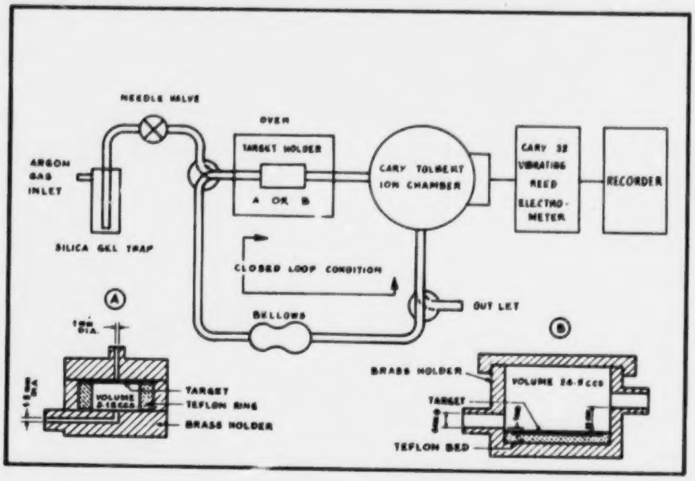
This paper deals with the studies on the trends of tritium released from used and fresh Ti-T sources under three different conditions, viz., dynamic equilibrium of the target with its surroundings, constant leakage under different flow rates of carrier gas, and tritium loss at high temperatures.

Experimental Technique

The schematic of the experimental setup is given in Figure 1. The argon line includes a drying agent such as indicator silica gel and a fine control needle valve, previously calibrated. Two target holders (A and B) of different volumes are housed in an oven which has facilities for preheating the inlet gas. The oven also has a fine control knob for adjustment of very low flow rates.

Figure 1

Schematic of Experimental Arrangement.



The brass target holder A is similar to the one used in electron capture studies with gas chromatographs. With the help of teflon rings and neoprene washers, targets with center holes are mounted face down and leak-tight. The carrier gas enters from the bottom, coming into contact with the tritium source before pushing through the center hole of the target. Brass target holder B, of a slightly larger volume, has a teflon bed which houses the target facing upwards. The gas enters from the bottom inlet, which is approximately 1 mm distant from the target surface, and grazes it before the gas leaves the holder. A rubber bellows hand pump is provided for recirculation of the gas in the system.

The targets used were prepared by the Isotope Division, BARC, by the conventional method [7,8]. Titanium is vacuum deposited onto copper discs at high temperatures, and tritium from U-T traps is impregnated onto Ti at high temperature.

The three targets taken for analysis had different histories of use. Their initial source strengths were reported to be 3.7 Ci (target T-14), 300 mCi (target T-9), and 400 mCi (target T-5). Targets T-9 and T-5 are of the same area, 314.9 mm^2, while T-14 is 530.7 mm^2. Target T-9 and T-14 were used in a 5.5 MeV and 400 keV accelerator respectively. In addition, target T-9 has a history of exposure to various temperatures with different gas flow rates.

Experiments were conducted under three different conditions.

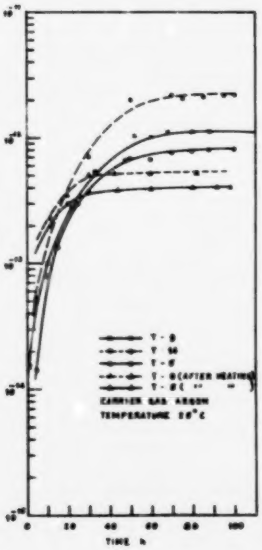
Closed-Loop Condition

The increases in chamber current using the three targets, with either of the target holders A or B, were measured at various time intervals. With continuous pumping with rubber bellows at constant intervals, the current gradually increases to a steady-state value after about four minutes of pumping.

For targets T-9 and T-5, the entire set of experiments was repeated with target holder A after a series of exposures to different gas flow rates, and at high temperatures (up to 200°C). The results are incorporated in Figure 2.

Figure 2

Build-up of Tritium Released from Target.

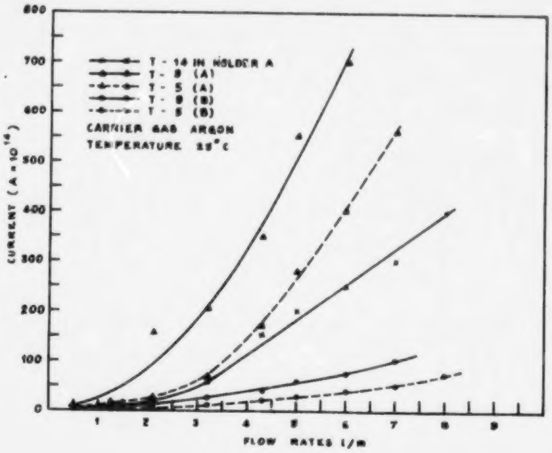


Once-Through Condition

This is done mainly to observe the behavior of the targets under a higher gas flow rate than is generally encountered in electron-capture studies. The tritium released from the target, as obtained from the steady-state values for each of the flow rates from 0.5 l/min to about 8 l/min, is measured using target holders A and B. The results are shown in Figure 3.

Figure 3

Variation of Released Tritium with Carrier Gas Flow.



Continuous Heating Under Constant Gas Flow

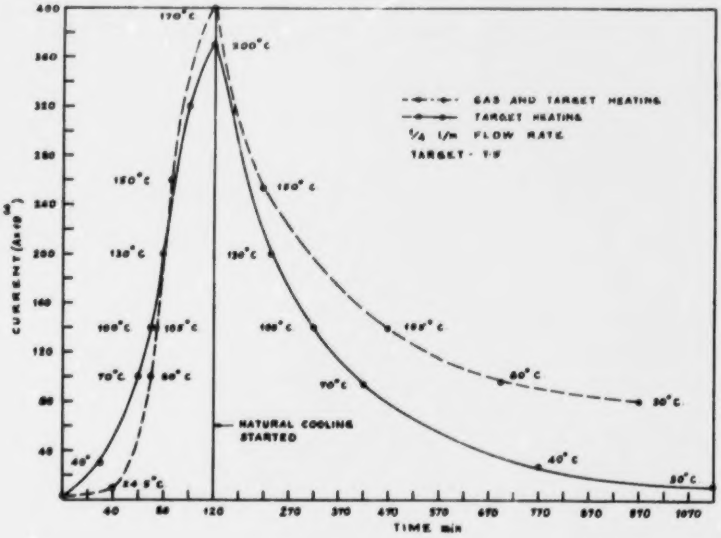
The gross tritium released from a heated target is monitored at different temperatures with a constant flow of gas. Results obtained with preheated gas passed over heated targets are also obtained using a very low flow rate, 0.25 l/min. Observations are repeated during natural cooling of the target. Figure 4 indicates the trend of the results obtained.

Results and Discussion

Figure 5 illustrates the normalized inverse build-up curve expressed as (Cm - Ct)/Cm vs. time (t). Notations Cm and Ct are tritium activities at equilibrium time and any time t, respectively. The values were obtained from Figure 2. The figure suggests that the curves, in general, can be represented by an expression of the type

Figure 4

Release of Tritium at High Temperature



$$\frac{Cm - Ct}{Cm} = A_1 + A_2e^{-\lambda_1 t} + A_3e^{-\lambda_2 t} \qquad (1)$$

where A_1, A_2 and A_3 are sample dependent constants and λ_1 and λ_2 are two rate constants characterizing the mode of desorption as measured in the system. One of these rate processes disappears after heating, as can be seen from the figure.

The curves were fitted using the method of least squares. However, the normalized inverse build-up curve for target T-14 before heat treatment does not seem to fit expression (1). The pattern of tritium activity as measured by the ionization current in the system can be seen from these curves. This is due to the desorption phenomenon, which may have more than one component depending upon its history of use. Desorption rates as envisaged in laboratory conditions are determined from the once-through condition, which takes into account all modes of tritium releases from the target.

Gross release rates continuously increase with gas flow rate for all three targets in the case of once-through condition. After each set of observations, the background of the system increased considerably, as shown in Table 1. Repeated flushing of the system with carrier gas restored the background to its original value; hence, the increase in

Table 1

Increase in Background Current (A x 10^{+15}) After Each Set of Once-Through Condition Observations

Target Holder	Target	Initial	Final	Background After Flushing
B	T-14	12	80	10
	T-5	8	100	10
	T-9	5	50	6
A	T-5	5	30	6
	T-9	5	12	5

background could be ascribed to an adsorption of tritium by the system walls.

The gross release rate K in μCi/h at various flow rates can be calculated as follows:

$$k = \frac{60 \; A_t \qquad f}{V} \qquad (2)$$

where A_t is the total tritium activity in the ionization chamber in μCi, f is the flow rate in l/min, and V is the volume of the chamber in liters.

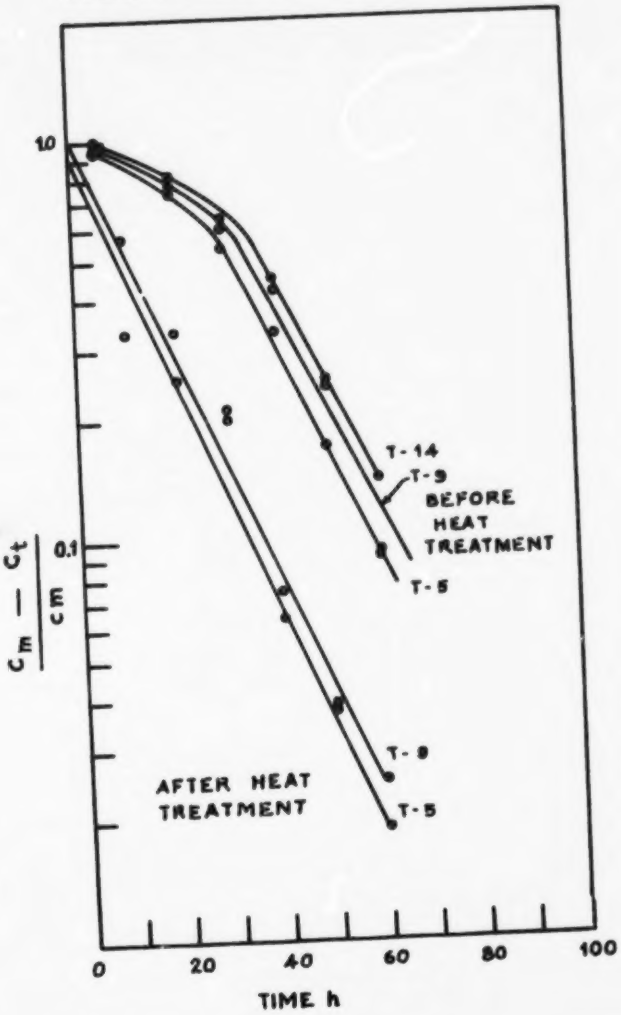
Table 2

Tritium Releases from the Target T-5 at Different Temperatures as Compared to Amersham Values

This Study			Amersham Values		
Target Strength	Temperature °C	Release Rate μCi/h	Target Strength	Temperature °C	Release Rate μCi/h
400 mCi	25	0.19-0.24	300 mCi	20	0.21
	80	3.31		100	3.5
	100	4.3		200	12.0
	150	8.6		250	135.0
	200	14.0			

Figure 5

Normalised Inverse Build-up of Tritium Activity.



Heating tends to increase the loss rate of tritium from target surfaces, see Figure 4 and Table 2. The values in Table 2 are computed during the cooling part of the experiment, when the temperature varies slowly with time. Table 2 also gives Amersham values [10] for comparison.

The present source strengths of the targets were checked by counting the bremsstrahlung from them, using a 1 mm thick and 2 inch diameter NaI crystal and counting gross X-rays. The present relative strength was calculated to be 1.46:0.4:0.29 as compared to the original values 3.7:0.4:0.3.

Conclusion

The kinetics of the desorption phenomenon from Ti-T sources have not been well explained. The measurements generally involve several unknown factors including adsorption by the various parts of the experimental system. The predictable behavior of a target with respect to its desorption rate depends entirely on the treatment it underwent during the impregnation and annealing [8,9]. Even under the best of conditions, there are unpredictable results, particularly the tendency of the target to flake off after tritiating [8]. Furthermore, there can be uneven surface distribution, due to the uneven evaporation of the metal in the first place [8].

From the closed-loop condition, it can be seen that a previously heated target kept in a closed container loses tritium through its inherent desorption phenomenon, the rate of which is quite low at room temperature. Experiments involving a large flow of carrier gas through the target can cause comparatively higher loss of tritium even at room temperature. This rate increases rapidly at higher temperatures. However, as long as the desorption phenomena involve the elemental form of tritium, the associated radiological hazard is limited. But if these situations lead to flaking off, the risk involved is quite high during preparation and use of the targets [11]. This is similar to abrading a target; tritiated dust or tritium powder of high concentration is very much more hazardous than tritium gas or tritiated water [8].

ACKNOWLEDGMENT

The authors are thankful to Dr. D. V. Gopinath for helpful discussions.

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THE CONTAINMENT OF ELEMENTAL TRITIUM AND TRITIATED WATER

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Operators of accelerator-type neutron generators using the deuterium-tritium reaction for neutron production recognize the difficulty of localizing their tritium inventory. Each target is a metal disc implanted with a few curies of tritium. Active programs may require an inventory of several tens of targets. Accordingly, the total inventory of tritium associated with an active accelerator program may run into the hundreds of curies. While compliance with appropriate standards for protection against radiation has not been difficult in storing and using multiple tritium targets, there has been difficulty in preventing the spread of tritium activity which can prejudice low-background counting. In addition, if an operator admits minors (those under 18 years of age) to his operation, it is less easy to demonstrate conclusively that permissible tritium concentrations in air have not been exceeded.

In light of this background, a work by Coffin [1] is of special interest to the users of tritiated accelerator targets or other substantial tritium sources. Coffin reported results of calculation which indicated that storage of tritium-releasing sources in plastic (polyethylene) containers would accelerate tritiated water production and, in general, aggravate the tritium control problem. Coffin's conclusion was that tritium-releasing materials would best be stored in well ventilated but secure areas, such as might be afforded by a closed wire basket. This work attempts to verify experimentally Coffin's analytically derived conclusions.

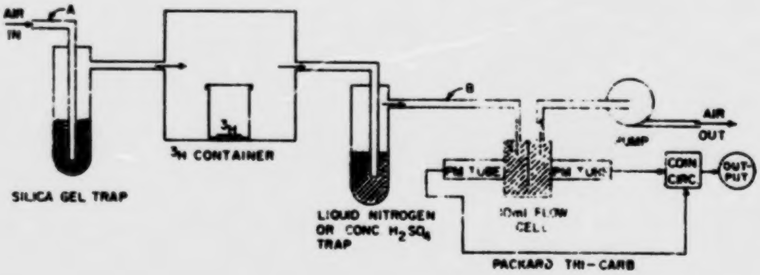
Experimental Procedure

Figure 1 is a diagram of the experimental apparatus used to study tritium containment by polyethylene. The volume of the outer tritium container was 3540 ml; the air volume flow rate at points A and B was 1440 ml/min. Therefore, the air in the container was exchanged once every 2.5 minutes.

The entire system, except for the pump, was enclosed in the refrigeration compartment of a Packard Tri-Carb liquid scintillation system. The inner tritium container was a 200-ml Nalgene beaker with a removable 2 mil polyethylene cover. The 10 ml flow cell was packed with anthracene, blue violet fluorescence, supplied by the Packard Instrument Co. Tritium sources used in this work were 3 to 5 Ci targets which had previously been mounted in a neutron generator. Provision was made to draw filtered and dried air through an enclosed volume into which elemental tritium could be metered. Provision can be made to place or remove a polyethelene barrier around the tritium source. Another water vapor trap is located downstream from the tritium source, followed by a flow cell in a liquid scintillation detector. Activation of the intermediate water vapor trap is required to remove tritiated water from, but leave elemental tritium in, the air stream.

Figure 1

Schematic of Experimental Apparatus Used to Study ^{3}H Containment.



A differing liquid scintillation detector response occurs following the emplacement of a tritium source as opposed to the placement of a plastic barrier. Change in response of the liquid scintillation detector to activation of the intermediate water vapor trap is an indication of the relative amount of elemental tritium in the air stream.

Figure 1 shows experimental apparatus used in this study. The five experimental regions considered are as follows:

I. Air was drawn through the system by a Gelman Little Giant Pump, Model 3152, to dry out the system and to get a baseline. This pumping lasted for one to two hours with no tritium targets present in the system. This air was pulled through a water vapor trap, e.g., liquid nitrogen, sulfuric acid, or silica gel.

II. The tritium target was placed in its container and simultaneously the liquid nitrogen or concentrated H_2SO_4 water vapor trap was removed.

III. When the scintillator response began to level out, the liquid nitrogen or concentrated H_2SO_4 water vapor trap was placed in the line.

IV. After one to two hours the polyethylene cover was placed over the beaker which contained the tritium target. The liquid nitrogen or concentrated H_2SO_4 water vapor trap was removed at the same time.

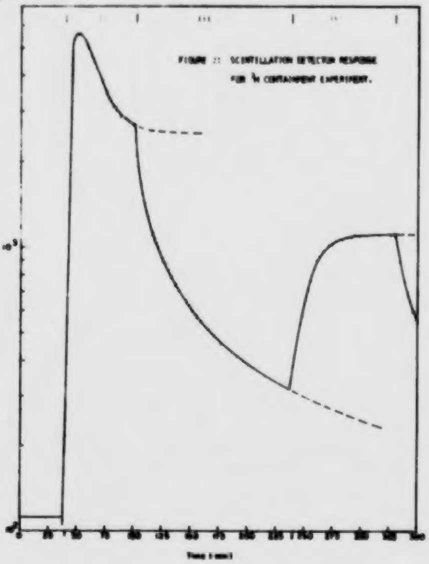
V. When the response leveled off, the water vapor trap was replaced.

Results and Discussion

Figure 2 shows the results of the experiments. Examination of the figure reveals several aspects of the experimental apparatus and procedures.

Figure 2

Scintillation Detector Response for ^3H Containment Experiment.



The peaking which occurred in Region II was probably caused by a buildup of tritiated water in the atmosphere above the tritium target inside the previously closed beaker.

Taking the characteristic times to be the time required for the detector response to reach an estimated 95% of the extrapolated equilibrium value, one obtains from Figure 2 the following characteristic times:

a. Region II - 100 minutes,
b. Region III - >250 minutes,
c. Region IV - ∿60 minutes.

The anthracene undergoes a "loading" phenomenon which occurs more rapidly than the unloading as is shown by the difference in characteristic times for Regions III and IV.

The response in Regions III and V indicates that most of the tritium is in the form of tritiated water.

The extrapolated asymptote for Region II has a value of approximately 2500 cpm (cpm is used here because only ratios of counts were used for this study, not absolute amounts that are usually indicated by units of dpm or μCi; all the work was performed on the same unit, thereby removing the necessity for converting units to an absolute unit). The saturation value for Region IV has a value of approximately 1100 cpm. Therefore, the polyethylene cover seems to reduce the overall emission of tritium by a factor of about 2.4.

This response, Figure 2, was typical of several experimental runs. There was up to ten percent deviation between the response shown in Figure 2 and the response of other runs. One run was made with a double-thickness polyethylene cover on the target beaker. Doubling this cover thickness had negligible effect on the system response in time or extent.

Conclusion

The plastic cover appears to provide significant containment of tritium that would otherwise be released to the surrounding atmosphere.

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TRITIUM LOSS FROM COATED CEMENT PASTE BLOCKS

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The concern with tritium oxide light water exchange arises from the consideration of the disposal of small quantities of lightly tritiated water obtained from the operation of the Los Alamos Meson Physics Facility (LAMPF) and other sources. These wastes could be contained in a casting made from Portland cement and tritiated water. Leaching, evaporation and diffusion of the tritium oxide from the porous castings could result in tritium exchange with environmental hydrogen and subsequent introduction of tritium into the biosphere. Accordingly, the development of an impermeable barrier in the form of a coating for the cement castings is essential to the success of a containment program based on the storage of tritiated cement paste solids.

In work to date, little more has been accomplished than a better definition of the parameters for a thorough study. Emphasis has been placed on use of the most economical coating materials, although no materials were eliminated from consideration on this basis.

Cement Paste Blocks

A strong, dense paste for the cement blocks was desired, hence a mixture with a water/cement (weight) ratio of 0.3 was selected. Variation in this ratio would affect a number of parameters of the movement of water in cement, such as porosity and rate of hydration. All cement pastes contain a certain amount of free water which increases as the water to cement ratio increases and varies with length of time of hydration.

The free, or evaporable, water in a mixture of water to cement of 0.3 (by weight) is about 50 percent of the total water in the paste. This was verified by heating a sample block for 70 hours at 100°C.

The selected mixture (0.3) provides a rather stiff paste. In certain applications, such as one which would require pumping, it is likely that the water to cement (weight) ratio would have to be increased to about 0.5, or additives would have to be introduced to provide a more fluid mix.

A Type I Portland general purpose cement was used for all samples. Retention of tritiated water in blocks prepared from other types of cement may differ somewhat, but this has not been investigated.

Experimental Procedures

Tritiated water which would provide 0.3 to 3 mCi of tritium per block of sample was used to form the paste. Cement and water were thoroughly mixed in a laboratory mixer for ten minutes, then spooned into glass or polyethylene wide-mouth vessels of 100 to 120 ml capacity. All samples were weighed at the time of forming and again just prior to coating. The weight difference due to moisture loss was noted and the accompanying tritium loss recognized.

The blocks were permitted to cure within their containers for a minimum of two days prior to removal and coating. In several cases, blocks remained within their molds for periods of up to a year before removal for coating application and study. In retrospect all samples should have been cured for the same period of time. However, the results would not be changed significantly.

Coating Materials

The first few groups of samples were coated to convenient and largely uncontrolled thicknesses with materials at hand. It was hoped that this would expedite the determination of which materials might be the most effective barriers to tritium.

Materials used in the preliminary investigation to coat the sample blocks included paving or road asphalt (Federal Specification SS-A-706c, grade AP-1, 120-150 penetration at 77°F); an epoxy-phenolic amine cured formulation, Plasite #1722 (Wisconsin Protective Coating Corp.); a universal quick-drying floor enamel (KOMAC, Denver, CO); a heavy duty resin-base organic solution for penetrating and sealing porous concrete, Peneclear (Penecrete Corp., Denver, CO); saran (vinylidene chloride); hypalon, a chlorosulfonated polyethylene elastomer; teflon "S" (tetrafluoroethylene fluorocarbon resin); heavy duty wheel bearing grease (Quaker State); steep (roofing) asphalt (200° to 215°F temperature range for softening point, minimum ductibility of 2.0 cm at 77°F, penetration between 1.5 and 3.0 mm at 77°F); Silastic 732, a room temperature vulcanizing (RTV) silicone rubber (General Electric); Glyptal, an air-drying or baking gloss enamel (principally rosin modified alkyd resin, aromatic and aliphatic hydrocarbons) designed for electrical resistance properties (General Electric); ceresin, a universal wax derived from purified ozocerite; beeswax; paraffin; and polyethylene.

Asphalt paraffin and beeswax, applied by dipping, formed coatings much thicker than the less viscous material such as floor enamel, Plasite, hypalon (spray), teflon "S" or Glyptal and were expected to be superior in retaining tritium. However, other of the more viscous materials such as grease or Silastic 732 were noticeably ineffective even at considerable thicknesses.

Several series of blocks were coated with varying thicknesses of asphalt, wax and paraffin. The technique for forming the thicker coatings consisted of dipping the block into the hot material for a few seconds, removing it and permitting it to set, then repeating this cycle until the desired thickness was achieved.

Certain coatings were used which were not applied by dipping because of the high temperature of the material in the liquid state. In the case of saran and polyethylene, the blocks were wrapped and sealed in thin films of the material. In another instance, a cement-paste-filled polyethylene container was sealed and the container itself was considered as a coating.

The quantity of coating applied is easily determined by weighing the sample before and after coating. Determination of thickness, however, is much more difficult and, in these tests, it frequently was not uniform. Suggested methods for thickness measurement included direct measurement using micrometer calipers, the eddy current method, the beta probe and radiography. The eddy current equipment requires that the specimen be conductive; the beta probe uses a special contacting device which is not particularly suited to the shape of the sample. Radiography, however, was successful in graphically illustrating coating thickness on X-ray photographic plates. With certain coatings such as Plasite, thicknesses less than about 0.127 to 0.178 mm were not detectable although a 0.20 mm coating was measured along the bottom of the sample. The first steep asphalt coated specimen showed a coating thickness of 2.41 mm on the lower half of a side on which the coating was typically 0.89 to 1.14 mm. The measurements confirmed the nonuniform coating conditions and served to support revised coating procedures in the latest tests.

Examination of Exchange Phenomena

The purpose of leach, or in this case "exchange," testing of radioactive solids is to determine the rate of loss of the isotope to its environment. Where encapsulated or "fixed" wastes are buried, the most serious consequences of this loss are related to contamination of ground water. The testing is conducted, therefore, in a water environment. In these experiments, the cement paste blocks were placed in 500 ml of demineralized water in a 0.95-liter polyethylene freezer container. The blocks with thick asphalt coatings were immersed in 1.5 liters of water in a four-liter polyethylene container.

The water, in all cases, was changed and samples were collected at weekly intervals for a short period and thereafter at regular four week intervals. The tritium content of the collected samples was determined through use of an organic-liquid scintillator using 0.5 ml of sample in a 12 ml cocktail. The cocktail, Aqua-sol, uses a xylene solvent with naphthalene for removal of quenching, a solute and two wavelength shifter phosphors. Normally the samples were counted for 100 minutes and the efficiency was about 30 percent.

One uncoated block was allowed to dry in an open hood to determine the rate of loss of tritiated water to an air environment. Twice each week the sample was placed in a sealed vessel for periods of one-half to six hours, the atmosphere of the vessel was transferred to an evacuated ionization chamber and tritium activity was determined. The data, which indicate a rather high initial rate of loss, are provided in Table 1.

The mechanism by which tritium is released from a material is complex, and in many cases may be a combination of several processes [1]. Movement through a coating may occur by diffusion through microscopic openings, or by tritium exchange of the tritiated water with hydrogen of the free and combined water in the cement paste or of the coating itself. Diffusion plays a major role with porosity affecting its rate to a high degree.

Methods of preventing escape of tritium to the environment would include fixing it into a compound which will not permit exchange or diffusion of hydrogen or water, or coating a tritiated cement paste with such a compound. The first may be possible but the method has not been investigated in this study. For the second suggestion, it is likely that in certain of the plastics, hydrogen is tightly bound and will not exchange and the materials are impervious to moisture. The study of plastics is being expanded.

Other substances, such as beeswax, contain tightly bound hydrogen

Table 1

Tritium Release from an Uncoated Block in Air

Total activity in block - 2.05 mCi

Days after Removal from Container	Tritium Release Rate %/sec	Calculated Cumulative Percent Release of Tritium
1	3.27×10^{-5}	1.4
5	5.02×10^{-6}	4.7
8	2.68×10^{-6}	5.4
12	1.61×10^{-6}	5.8
15	8.49×10^{-7}	6.4
19	1.54×10^{-6}	6.6
22	5.95×10^{-7}	6.8
26	9.32×10^{-7}	6.9
28	4.10×10^{-7}	7.0

and are very effective barriers. It is possible that materials which contain no hydrogen may prove superior. None of these hydrogen-free materials have been tested as yet.

Tritium Release Rates from Coated or Uncoated Blocks

In this paper, release or loss rate refers to the percentage of the total quantity of tritium initially included in the sample block which is found in its water environment after a certain period.

Three series of cement paste samples were mixed with the following tritium contents:

Series "T" - About 0.32 mCi per block

Series "H" - About 3.0 mCi per block

Series "W" - About 2.3 mCi per block

Figures 1 and 2 illustrate the increase in percentage released over a period of time. The uncoated control samples, of course, release or lose tritium most readily. The first control sample of the "T" series has lost about 60 percent of its tritium content in 72 weeks, with half of this amount being lost in the first six weeks.

Figure 1

Comparative Release of Tritium from Coated and Uncoated Tritiated Cement Paste Blocks Immersed in Water

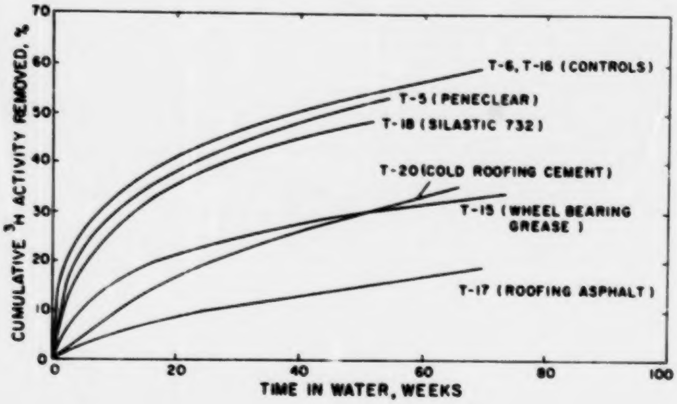
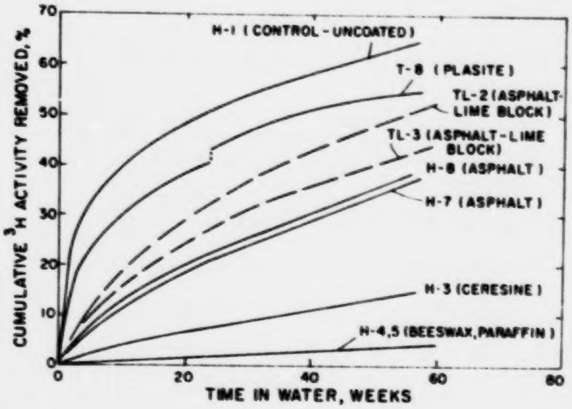


Figure 2

Comparative Release of Tritium from Coated and Uncoated Tritiated Cement Paste Blocks Immersed in Water Including Data for Two Asphalt-tritiated Lime Blocks



Many of the curves indicate this rapid initial loss. With the more effective barriers such as asphalt, the cumulative release curve is nearly linear, indicating a rather uniform release rate throughout the test.

The beeswax and paraffin curves indicate that these materials provide the most effective barriers. It must be pointed out, however, that for the samples indicated in Figures 1 and 2, samples H-4 and H-5 are among those with the thickest coatings. The asphalt coatings, other than H-7 and H-8 which were immersed in hot asphalt for extended periods, are relatively effective barriers and the use of asphalt is being studied in some detail.

Figure 3 shows the fractional release per week of residual tritium as a function of immersion time. The figure indicates that the exchange or release mechanism is complex and cannot be clearly defined at this time.

Figure 3

Fractional Release per Week of Residual Tritium
as a Function of Immersion Time

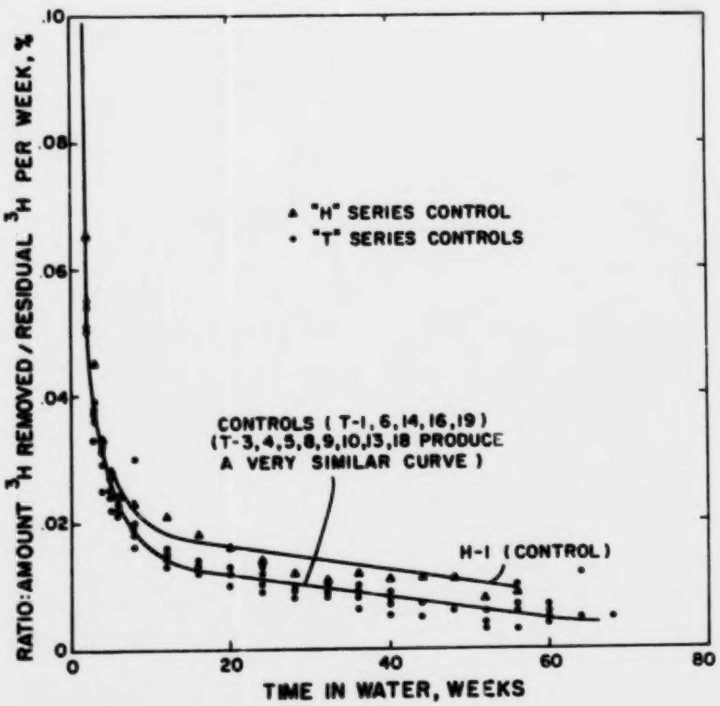
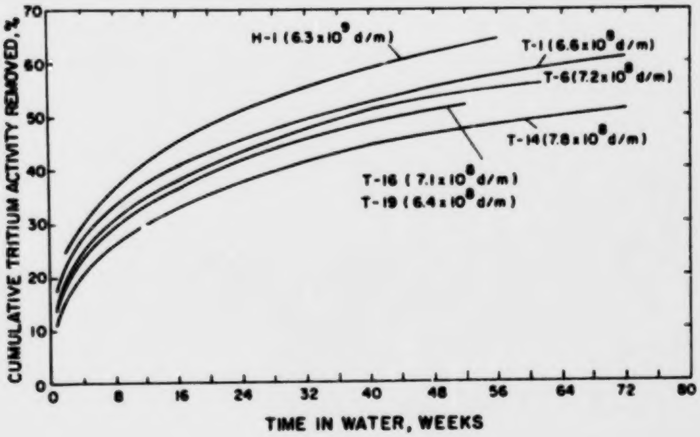


Figure 4 provides cumulative removal curves for a number of control samples. The curves are very similar, indicating a high initial loss of tritium which decreases rather uniformly. Samples T-1, T-6, T-14 and T-19 are all from the same cement-tritiated water batch, but they were immersed in water for the first time at successive one week intervals in the order stated. It would appear that the older samples with a higher percentage of bound water should have a lower loss rate which is the case for all except the T-14 curve, for which no explanation is offered.

Figure 4

Comparative Release of Tritium from Uncoated
Tritiated Cement Paste Blocks Immersed in Water



In later attempts to provide asphalt coatings of greater thickness, three cement paste block samples were dipped for very brief periods in steep asphalt which was heated to about 150°C. The samples were air-dried for 15 seconds between dippings. Coating weights which resulted varied from 40 to 45 g. These samples were prepared only recently and the loss or release data available at this time for a typical sample are shown in Table 2.

Three samples were placed in paper cartons and surrounded with steep asphalt to provide a 2.5-cm thick coating at top and bottom and a circumferential coating thickness of about 1.9 cm. These "cast"

Table 2

Release Rates of Tritium
Through an Asphalt Coating

Immersion Time, Days	"Dipped" Block Cumulative % Tritium Released	"Cast" Block Cumulative % Tritium Released
7	0.08	0.0040
14	0.17	0.0061
21	0.26	0.0075
28	0.36	0.0086
35	0.48	--
42	0.60	--
56	0.80	--

blocks were immersed in 1500 ml of distilled water and early data indicate that this thick coating is very effective in containing the tritium, with release rates essentially zero as indicated in Table 2. Coatings of 2.5 cm of asphalt would be neither difficult to apply nor prohibitive in cost. Beeswax or paraffin of similar thickness may well be as effective and will be tested, but they would be much more expensive to use in actual practice. A similar argument can be used with respect to polyethylene.

Four samples were encased in sealed polyethylene bags and from 0.08 to 0.7 percent of the tritium was released during the first week. Though much heavier polyethylene, which is being tested, may prove as satisfactory as the thick asphalt, it is unlikely that the cost of the polyethelene will compare favorably in the near future.

The chemical and physical characteristics of the matrix have a major role in control of the variation in release rate. Loss from a matrix other than cement is shown in the plot of the special asphalt samples in Figure 2. These special samples, TL-2, TL-3 and TL-4, were prepared by slaking quicklime with tritiated water in stoichiometric amounts and mixing this lime with steep asphalt. The matrix in this case is the much less porous asphalt which contains no readily evaporable water and which appears to lose tritium at a much more uniform rate throughout the period than the uncoated cement paste blocks.

Conclusions

Though the study is in its early stages, it has shown that a tritiated water-cement paste mixture provides a solid from which the tritium is released readily into an air or water environment. Several coatings have been investigated, which, in sufficient thickness, act as a reasonably effective barrier to the loss of moisture and the release of tritium. These materials, asphalt, paraffin and beeswax, are relatively common and the first two mentioned are available in large quantities at reasonable cost. For large accelerators, power reactors or fuel reprocessing plants, the cost of tritium disposal with this process would be minor even with very expensive coatings.

Continued investigation may uncover more effective coating materials or, ideally, a compound into which tritium may be incorporated in a nonlabile form by chemical reaction with dilute tritiated water. A thorough, more sustained study is necessary and it is hoped that this paper will stimulate other efforts along this line.

ACKNOWLEDGMENTS

The authors are indebted to Mr. Patricio Trujillo who analyzed most of the water samples for tritium. The work was done under the auspices of the U. S. Atomic Energy Commission.

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DETERMINATION OF TRITIUM LEAK RATES FROM WASTE PROCESSING EQUIPMENT

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The procedures described in this report were developed in support of the Mound Laboratory program for recovery of tritium from waste materials. Tritium is notoriously difficult to contain, and it is of vital importance from an ecological standpoint that there be no appreciable leakage from containers in which tritiated materials are shipped or stored. After processing, wastes must be put into cannisters which are welded shut, and these must be transported from monitored areas to burial sites. Oil in the vacuum pumps used in the recovery process must be changed and safely contained. In addition, it is sometimes useful to be able to test a prototype of a component of the recovery system, such as an O-ring seal, for leakage, since part of the system is operated at 3 atm pressure. Analytical techniques were therefore developed for determining leak rates of 10^{-16} ml/sec and higher.

The method involves sealing the cannister, containing tritium wastes, in a methane atmosphere within an appropriate size container, the interior of which has been coated with cellulose acetate. After several days, the tritium which has leaked from the cannister into the container is measured by beta counting.

Adsorption on the sides of containers, vacuum lines, counting tubes, etc., is a serious problem in the measurement of tritium in gas, particularly when moisture or oil vapors are present [1], or when long exposure to tritium occurs. For this reason, cannisters are sealed in inexpensive containers which can be discarded after use, so that tritium from one measurement will not influence the next when desorption may occur. In addition, the inside of each container is coated with cellulose acetate before it is sealed, and after the tritium has been collected, the cellulose acetate is peeled off and dissolved. The adsorbed tritium is then measured by liquid scintillation counting. Tritium in the methane is measured by gas proportional counting. The total tritium collected is the sum of the adsorbed tritium and the tritium in the methane gas.

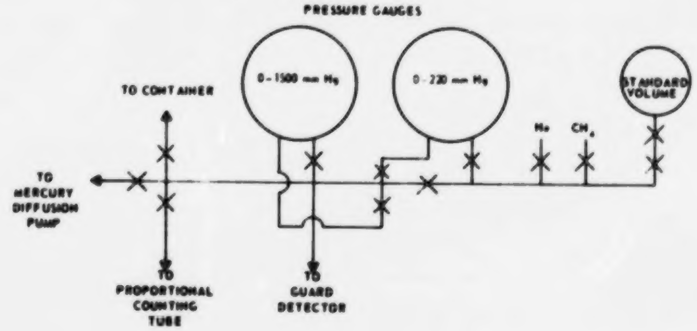
Collection Procedure

Disposable containers are used, in which cannisters are sealed and tritium is collected. One- and five-gallon cans (Freund Can Co., Chicago, IL), and 100-pound "lard" cans (Heekin Can Co., Cincinnati, OH), are used to accommodate the various size vessels and prototypes of equipment used in the processing and storage of tritium waste. A "quick disconnect" Swagelok coupling is attached to each side of the container. The one-gallon can is mechanically sealed. The seams of the larger cans have to be soldered, but with this precaution and with the aid of lacquer and sealing putty, the containers are made airtight. The inside of the can is coated before sealing with cellulose acetate replicating tape (Ernest F. Fullam, Inc., Schenectady, NY) dissolved in acetone to a 7% solution. Three coats of the solution are applied with a paint brush, and each coat is dried with heat lamps.

After the cannister is sealed inside the coated container, the container is connected to the vacuum system by the "quick disconnect" coupling. A diagram of the vacuum system is shown in Figure 1. The

Figure 1

Vacuum line system



containers would collapse if they were evacuated, so the air is removed from the container by flushing with helium, and the helium is then displaced with methane. The valve to the container is closed, and the time and pressure (slightly above atmospheric) are recorded.

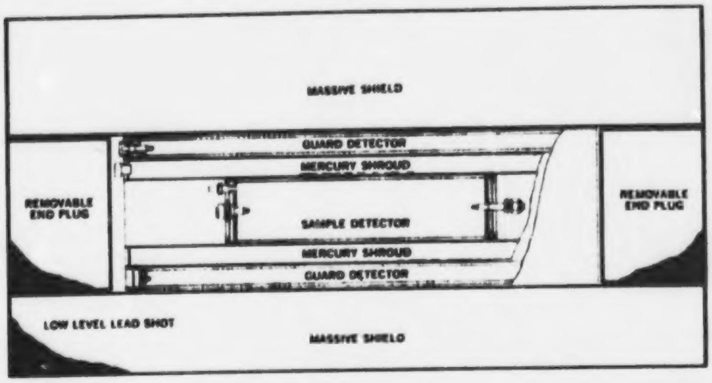
After two or more days, depending on the magnitude of the leak rate and the size of the collection chamber, the accumulated tritium is measured. The valve to the collection chamber is opened, and a portion of the methane gas is expanded either into a one-liter flash, or, depending upon the volume of the collection chamber, simultaneously into a one-liter flask and directly into the proportional counting tube. The gas in the proportional tube is counted, and aliquots of the gas in the one-liter flask are sampled for additional measurements.

Proportional Gas Counting

The detection of tritium by gas proportional counting [1] is a fairly common method of detecting this isotope in gaseous samples. The proportional tube and shielding system are shown in Figure 2. In order to reduce the background within the sample detector, a guard detector proportional counting tube surrounds the sample detector and, with associated electronic equipment, acts as anti-coincidence shielding [1]. The sample detector and guard detector are placed inside a lead shield about 15 cm thick. A diagram of the electronics system is shown in Figure 3. The gas detector system with shielding was manufactured by Radiochemistry, Inc., the preamplifiers by Radiation Instrument Development Co. and the remainder of the electronic equipment by Baird Atomic.

Figure 2

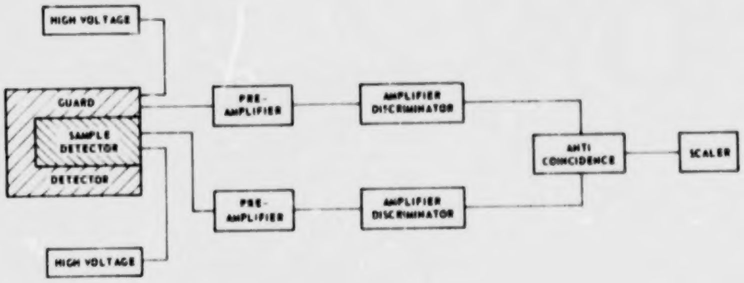
Proportional tube and shielding system



To determine the operating potential of the gas proportional counting tube, it is necessary to determine a voltage plateau with methane as the counting gas, since the tritium is collected in a methane atmosphere. The tritium could be collected in air or in certain other gases, but methane is used because it is an ideal proportional counting gas. Voltage plateaus at 50 cm Hg pressure are illustrated in Figure 4. For leak rate determinations, the plateau is determined with an external source if the counting rate of the sample alone is too low to afford good statistics. A 10 μCi cesium-137 sample is used for this purpose. The source is placed adjacent to the sample detector, and the Compton electrons from the gamma ray interactions within the sample detector serve as a source of "beta" particles.

Figure 3

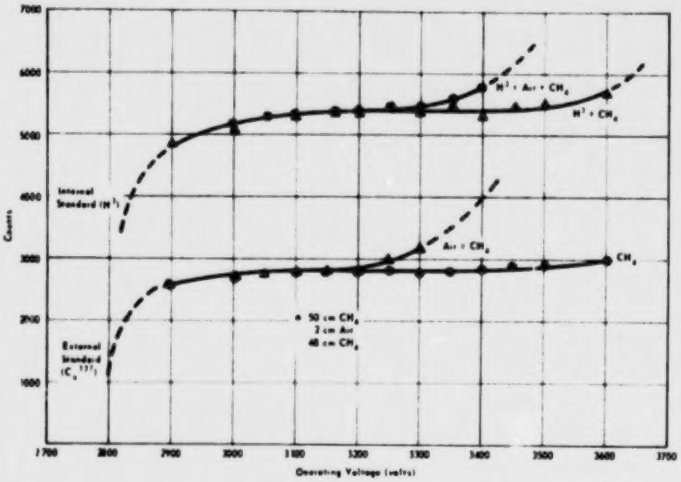
Block diagram of detection electronics system



The effect of the presence of air in the proportional counting tube has been examined, since occasionally one of the cans leaks. The lower curves in Figure 4 were observed with pure methane in the counting tube and with methane plus 4 vol % air, using an external source. The upper curves were prepared by diluting a tritium in helium-3 sample 100 to 1 in pure methane, and diluting the same tritium in helium-3 sample by the same factor in methane plus 4 vol % air. The curves indicate that the voltage plateaus are shorter when the gas contains air, and that the proper operating voltage is about 50 V below the upper end of the plateau determined with an external source. This voltage will be approximately at the middle of the plateau of the sample alone. The curves also demonstrate that tritium measurements are valid with this concentration of air in methane, provided the proper operating voltage is used.

Figure 4

Voltage Plateaus



Measurement of Adsorbed Tritium

Immediately after the gas is expanded from the collection chamber into the counting tube and flask, the can is opened and the cellulose acetate is peeled from the interior with forceps. Segments of the coating removed from the small cans are divided into about 10 portions, and each portion is placed in a 25-ml polyethylene scintillation counting vial. Twenty milliliters of scintillator solution, prepared from 14 g 2,5-diphenyloxazole (PPO), 200 g naphthalene, and 100 mg 1,4-bis[2-(5-phenyloxazolyl)] benzene (POPOP) diluted to two liters with 1,4-dioxane, is added to each vial. After the segments have dissolved in the scintillator solution, the samples are counted. The coating peeled from the five-gallon can or 100-pound "lard" can is placed in a 500-ml or 1000-ml volumetric flask, and scintillator solution is added, to volumes of 500 ml or 1000 ml, respectively. After the coating has dissolved, six 20-ml aliquots of each are measured by liquid scintillation counting. The counting efficiency is determined for each sample by automatic external standardization. A Packard Tri-Carb Liquid Scintillation Spectrometer is used for scintillation counting.

The measurement could be affected by tritium adsorbed on surfaces such as the vacuum lines, the sample flask, and the counting tube. To avoid this, flasks are rinsed with acetone and then evacuated to assure a clean, dry surface to which tritium does not readily adhere during the short time it is left in the flask. After a leak test has been completed, the counting tube and flask are evacuated, filled with methane and allowed to stand overnight, after which background measurements are made.

Past experience has indicated that adsorbed tritium is not effectively removed by pumping, but that the tritium does exchange with hydrogen in the methane and is then detected. The amount of tritium adsorbed on the sample flasks after leak tests have been completed is insignificant compared to the tritium found in the acetate tape which has been in contact with the tritium over an extended period of time on the interior of the collection chambers.

Calculations

The disintegration rate (D_t) of the total tritium collected in the gas is related to the fraction counted as follows:

$$D_t = \left(\frac{P_t V_t}{P_a V_a}\right) D_a$$

where P_t = pressure of gas in the collection chamber,
V_t = volume of gas in the collection chamber,
P_a = pressure of sample gas in the counting tube,
V_a = effective volume of the counting tube, and
D_a = disintegration rate of the aliquot counted.

The effective volume of the counting tube, V_a, is calculated from the dimensions of the tube by

$$V_a = \pi r^2 h$$

where r = radius of the tube and
h = length of anode wire within the counting tube.

The disintegration rate, D_s, of the total tritium adsorbed on the interior of the can is given by:

$$D_s = \frac{V_c}{20n} \sum \frac{C_i - C_b}{E_i}$$

where V_c = total volume in ml of the dissolved cellulose acetate segments,
n = number of 20 ml aliquots,
C_i = cpm of each aliquot,
C_b = cpm of background, and
E_i = counting efficiency for each individual sample. This is the ratio of counting rate to disintegration rate.
(For the one-gallon can, $\frac{V_c}{20n} = 1$)

The overall leak rate of tritium, R, in ml/sec is

$$R = \frac{V}{t}$$

where V = volume in ml of collected tritium, and
t = collection time in seconds.

The volume of tritium collected is determined from the disintegration rate of the gaseous and adsorbed tritium by

$$V = \frac{D_s + D_t}{5.32 \text{ X } 10^{12}}$$

where 5.32 X 10^{12} = specific activity of tritium gas in dis/min·ml at 22°C, and 1 atmosphere pressure.

Sensitivity

The sensitivity of the measurements depends in part upon the size of the cannister whose leak rate is to be determined, and hence on the size of the collection chamber required. For a cannister sealed in either a 100-pound "lard" can or a five-gallon can, and a collection time of one week, leak rates approaching 1 X 10^{-16} ml/sec may be detected. For smaller cannisters sealed in a one-gallon can for one week, leak rates of 2 X 10^{-17} ml/sec may be detected. These limits represent gas counting rates equal to the square root of the background, the background being about 60 cpm for a two-liter copper gas proportional counting tube. Gas to 50 cm Hg pressure may be expanded directly into the counting tube from 100-pound cans, to 25 cm Hg pressure from five-gallon cans, and to 20 cm Hg pressure from one-gallon cans, without causing the cans to collapse.

For example, methane containing tritium leaking from a cannister in a 100-pound "lard" can for seven days, expanded into a counting tube to 50 cm Hg pressure, gave 8 cpm over background. No activity was detected on the cellulose acetate. Data and calculations for this determination are as follows:

P_t = 76 cm
V_t = 6 X 10^4 ml
P_a = 50 cm
D_s = 0

V_a = 1700 ml
D_a = 8 cpm
t = 7d = 6.05 X 10^5 sec

$$D_t = \frac{P_t V_t D_a}{P_a V_a} = \frac{(76)\ (6 \text{ X } 10^4)8}{(50)\ (1.70 \text{ X } 10^3)} = 429 \text{ dis/min}$$

$$R = \frac{V}{t} = \frac{D_s + D_t}{(5.32 \text{ X } 10^{12})t} = \frac{0 + 429}{(5.32 \text{ X } 10^{12})\ (6.05 \text{ X } 10^6)} =$$

$$1.3 \text{ X } 10^{-16} \text{ ml/sec.}$$

Higher leak rates can, of course, be measured with greater certainty.

Validity of the Measurements

The tritium gas counting techniques and the method for determining the adsorbed tritium were evaluated to establish the validity of the measurements. To evaluate the gas counting techniques, tritiated hydrogen gas was prepared by the reduction of standardized tritiated water. The absolute disintegration rate of the water was determined by liquid scintillation counting using a National Bureau of Standards (NBS) tritiated water standard. After reduction of the tritiated water with magnesium, the hydrogen gas was diluted with methane, and its activity was calculated. The activities of five such samples are given in column two of Table 1. These samples were counted in a two-liter copper proportional counting tube and in a 500-ml Pyrex tube. As indicated by the results in Table 1, good agreement was observed between the activities measured by gas proportional counting, and the values calculated on the basis of the NBS standard tritiated water. The low activity on the third sample is probably due to the fact that some of the tritium adsorbed on the flask in which it was stored for 2.5 days before it was counted. Residual activity was found in the flask after it was evacuated and refilled with methane. The other four gas samples were measured immediately after reduction and dissolution, and no residual activity was found in the flasks which had contained them.

The volumes of the vacuum lines and flasks were determined by comparison with a one-liter round-bottom Pyrex flask, whose volume was determined with an accuracy of ±0.1% in comparison to calibrated laboratory standards traceable to the NBS. The volumes of gas in the collection cans are determined by measuring the volume of water required to fill them and subtracting the volume of water displaced when the cannister to be measured is placed in the can. If the cannister cannot be immersed in water, or if its volume cannot be determined accurately by measurement of the physical dimensions, the volumes of gas in the collection cans is determined by pressure difference before and after expansion into a calibrated flask. The second method is less desirable, since the sides of the cans are not entirely rigid, and small volume changes occur when the pressure changes.

Table 1

Gas Proportional Counting of Tritiated Hydrogen Prepared From
NBS Standardized Tritiated Water

Sample Number	Tritium Content in dis/min·ml X 10^4		Deviation, Gas Proportional Counting From NBS Value
	NBS Value	Gas Proportional Counting	
1	2.91	2.96	+1.7%
2	2.37	2.36	-0.4%
3	1.92	1.76*	-8.1%
4	2.26	2.27	+0.4%
5	2.79	2.74	-1.8%

*Low proportional count due to adsorption. See text.

Other possible errors include those due to counting statistics, errors introducted into gas counting if acetone fumes from the cellulose acetate or vapors from the lacquer used as a sealant remain in the cans, air remaining in the cans because they are insufficiently flushed, or cans leaking so that the methane in them partially exchanges with the room atmosphere. These errors are avoided when possible, and all measurements are made in duplicate or triplicate to give added reliability to the results.

The overall accuracy of the leak rate determinations was evaluated experimentally in the following manner: three one-gallon and three 100-pound "lard" cans were coated with cellulose acetate, sealed and flushed with helium and then methane, as for a routine leak test. Each can was partially evacuated, and a known amount of tritium in methane was expanded from a one-liter flask into the can. The amount of tritium introduced was determined from the concentration of tritium in the flask and from the volumes and pressure difference in the flask and vacuum lines before and after expansion into the can. The pressure in the can was brought to slightly above atmospheric with methane and the can was sealed. After one day or longer, the tritium in the gas and on the cellulose acetate was measured, and the amount of tritium recovered was compared to the amount introduced. Results listed in Table 2 show that recovery was complete.

Table 2

Recovery of Tritium From Collection Chambers

Test Number	Size Can	Tritium into Can dis/min x 10^6	Time (h)		Tritium Recovered dis/min x 10^6		Percent Recovered
1	1 gallon	8.74	[illegible]		8.79	in gas	
					0.00	on tape	100.6
				Total	8.79		
2	1 gallon	10.67	64.3		10.54	in gas	
					0.03	on tape	99.1
				Total	10.57		
3	1 gallon	4.05	20		3.90	in gas	
					0.046	on tape*	97.4
				Total	3.946		
4	100 pound	78.3	92		78.1	in gas	
					0.7	on tape	101.1
				Total	78.8		
5	100 pound	149.8	7[illegible]		150.1	in gas	
					000.0	on tape	100.[illegible]
				Total	150.1		
6	100 pound	[illegible]	[illegible]		[illegible]	in gas	
					000.0	on tape	101.7
				Total	[illegible]		
							Average 100.[illegible] ± [illegible] (standard deviation)

*Air was known to be present in this can

ACKNOWLEDGMENTS

The authors acknowledge with gratitude the contribution of R. K. Gillette who prepared the hydrogen samples from standardized tritiated water. Mound Laboratory is operated by Monsanto Research Corporation for the U.S. Atomic Energy Commission under Contract No. AT-33-1GEN-53.

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* * *

EXPERIENCE IN RECONDITIONING ION PUMPS CONTAMINATED BY TRITIUM DURING NEUTRON GENERATOR USE

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It has been estimated [1] that about 200 neutron generators are in use in the United States today, mostly of the Cockroft-Walton type. Ion pumps are used to generate the ultra-high vacuum (10^{-8} mm Hg pressure or lower) required by these generators. The ion pump description and theory of operation are found in the literature [2]. These generators operate by accelerating a beam of deuterium ions at energies up to approximately 200 keV into titanium-tritium targets. The d-t reaction produces the 14 MeV neutrons. The targets, however, become inoperable after a few hours' use by the deuterium exchanging for the tritium, thus necessitating frequent reloading with new targets. Each target contains about 0.6 Ci T/cm^2. Estimates have been made [3], indicating as much as 40 to 50 curies of tritium build-up in the ion getter type pumps after the use of 20 to 30 conventional targets of 0.5 to 0.8 Ci/cm^2.

Various surfaces and loose particles in the pump interior become tritiated with use. The particles found in contaminated pumps range in size from fine powder, in the submicron size range, to large pieces of scale. All of these tritiated parts emit tritium at a measurable rate into the pump interior. Engleke [4] discussed the health problems associated with a Van De Graaf accelerator. Work at this laboratory indicated that it was neither feasible nor practical to remove all tritium contamination from a pump during reconditioning because the pump immediately becomes contaminated when reinstalled on an accelerator. A reconditioning procedure was established to restore the pump to an operable state. This is done by cleaning the pump interior, removing loose tritium-contaminated particles and replacing the "getter" elements.

Revamping Procedures

At the generator facility, the contaminated ion pump is carefully demounted from the generator under well-ventilated conditions, taking special care to avoid blowing the potentially hazardous particulate material into the working area. Flanges, supplied by the manufacturers, are used to seal the ends of the ion pump. The magnets are removed and the pump body, wrapped in plastic, is crated in a suitable wooden container for shipping. It is affixed with a radioactive white Class 1 label, or as determined by the transport index [5].

The pump, as received at the reconditioning facility, is wipe-tested for gross activity and tritium prior to service handling. Proper protective clothing is worn by personnel during the processing. The pump is placed in a well-ventilated hood (38 m/min) with absolute filters. A calibrated tritium gas monitor is operated during the removal of the contaminated Penning cells, which are transferred to plastic bags and sealed in tin cans for ultimate disposal. Most of the particulate material is removed with dampened disposable wipes, and remaining traces of particles are removed by washing. The pump body is filled with water, agitated, and the contaminated rinses siphoned into collection bottles. The liquid waste is assayed by liquid scintillation counting to determine the proper dilution factors required for disposal. Washings are repeated until negligible particulate material remains. The interior of the pump is dried, new Penning cells are installed, and the flanges are replaced. The exterior of the pump is checked for tritium contamination prior to return of the revamped pump. If a swipe over a 100-cm^2 area measures higher than 50 dpm, the pump body is thoroughly wiped clean with dampened disposable wipes until contamination is below 50 dpm. Tritium urinalyses are performed on personnel involved in pump reconditioning.

Monitoring Tritium Contents of Pump Interior

This laboratory uses a Tracerlab MGP-3 radioactive gas monitor [6] which is calibrated with known amounts of tritium delivered directly from previously calibrated holding tanks into the sniffer nozzle. A 100 MCi $^3H_2$ source, diluted with air and pressurized, is used for the calibration. Various amounts of $^3H_2$ are delivered to the sniffer and the detector response is recorded on strip chart paper. Calibration is obtained for all three scale readings after the detector zeroing sequence is performed. The results showed good lineararity and reproducibility at the X 10 and X 100 scale settings. The monitor is capable of detecting from 3.7 X 10^{-5} μCi/ml to 1.4 X 10^{-3} μCi/ml. This compares well with the MPC_a working area limit of 5 X 10^{-6} μCi/ml [7].

Some representative results from the reconditioning operations, as recorded by the gas monitor, are shown in Table 1. Results vary widely from pump to pump. These measurements were obtained from various contaminated pumps by placing the sniffer directly over the flange opening immediately after the flange cover was removed. Pumps shipped to the laboratory were all found to be so contaminated that, when opened for the first time, they drove the monitor off scale. The tritium activity comes from the outgassing of the Penning cells, the pump body, and the particulate material into the free spaces in the pump interior. During the monitoring measurement, this activity is swept out of the pump body in less than one minute. The activity build-up can easily be repeated by replacing the flange cover for a period of time and heating the pump externally with heat lamps.

The data indicate large amounts of tritium remain in the pump body walls even after removal of the Penning cells (elements) and particles. The tritium outgassing appears to be quite rapid. With a rather drastic chemical treatment ($KMnO_4$ and citrate solutions) as shown with pump C, the amount of tritium outgassing increased. This suggests that fresh tritiated surfaces were exposed in the pump body by the chemical treatment.

Particulate Material Found

The amount of particulate material varies from pump to pump and is

Table 1

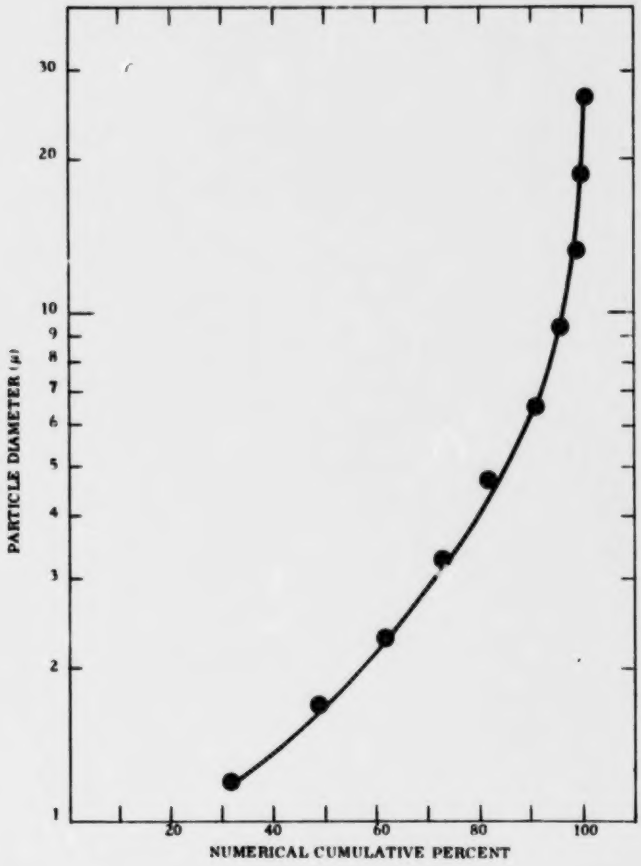
Range of Tritium Gas Levels Measured in Different Contaminated Pumps During Reconditioning

Activity Buildup Period (min)	Tritium Detected µCi/l	Average Rate of Buildup pCi/ml min	Remarks
		PUMP A	
120	2.6	22.0	Prior to reconditioning
10	0.53	53.0	Repeat
120	0.28	2.3	With elements removed plus heat
		PUMP C	
6	0.80	130.0	Prior to reconditioning
5	2.3	460.0	With elements removed
5	0.49	100.0	After particles removed
10	0.16	16.0	After baking pump body with heat lamp for 12 hours, flange off
150	0.60	4.0	Continue for four days, flange off
60	2.7	45.0	After rinsing pump interior with $KMnO_4$ and Citrate solutions and drying with heat lamp
		PUMP G	
72	7.5	100.0	Prior to reconditioning
300	8.7	29.0	Repeat
70	3.0	43.0	Elements and particles removed. Bake with heat lamp
155	3.0	19.0	Repeat particle removal and baking with heat lamp

apparently due to unknown accelerator operation parameters. The results of the particle size measurements of pump G are shown in Figure 1. The plot indicates that 97 percent of the particles have diameters less than 10 μ and 50 percent of the particles have diameters less than 1.8 μ. The results show the existence of a potential health hazard from lung deposition, in addition to a hazard to the digestive system through improper handling of opened pumps.

Figure 1

Numerical size distribution of particles from the interior of spent ion pump G contaminated with tritium.



Figures 2 and 3 are photomicrographs showing some typical irregularly shaped particles taken from the pump interior. All particles observed were flat and scale-like and variously colored, ranging from iridescent bluish black to reddish brown and even white or silver.

Figure 2

Photomicrograph of typical particles found from tritium contaminated ion pumps (x 100 magnification).

Figure 3

Photomicrograph of typical particles found from tritium contaminated ion pumps (x 250 magnification).

Microprobe Examination of Particulates from a Pump Interior

All of the pump bodies processed had a dark reddish brown to bluish black coating, of undetermined composition, throughout the interior of the pump. Some of this flashing or coating was easily removed from the high-voltage stainless steel straps taken from the spent pumps. It was then compared to the typical particulate material from the pump using an electron microprobe analyzer (Applied Research Laboratory electron-microwave model EMX-SM). The data from the X-ray intensity plots are compared in Figure 4. The results from this study demonstrate the identical nature of the two materials with the exception of a trace amount of iron found in the stainless steel strap sample. No explanation is given for the presence of the large amounts of tantalum.

Figure 4

Microprobe comparison scans of A material coated on stainless steel strap, and B typical particle from spent ion pumps.

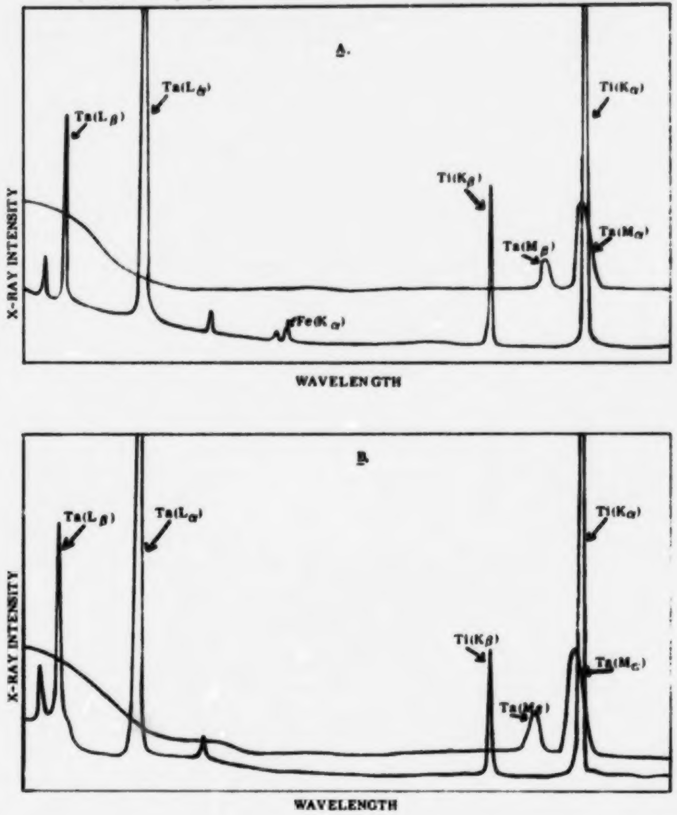


Table 2

Ranges of Tritium Levels Measured in Samples
From Different Contaminated Pumps

Sample Type	Tritium Detected	Remarks
1 ml rinse water nCi/ml samples	1.3	Pump A
	1700.0	Pump F
	100.0	Pump G
Wipes from spent nCi/100 cm^2 element Ti Cathode surfaces	4.5	Pump A
	1300.0	Pump G
Wipes from inside nCi/100 cm^2 walls of revamped pumps	36.0	Pump C
	1000.0	Pump F
	250.0	Pump G
Dry particles nCi/mg (10.2 mg sample)	4.8	Pump A
Bioassays of workers (urinalysis) pCi/ml	53.0	Pump A
	100.0	Pump F
	54.0	Pump G
	<14.0	Pre-exposure sample

Tritium in Samples from Contaminated Pumps

Samples were taken during various stages of pump reconditioning. Tritium in the samples was measured using liquid scintillation counting techniques. A Tracerlab Corumatic-100 liquid scintillation counter was used for the analyses. Results of representative samples are given in Table 2.

Tritium content of the samples varies widely from pump to pump. The data show that relatively high tritium values are associated with the contaminated pump parts and interior regions. The rinse water measurements, ranging from 1 nCi/ml on up, are much more contaminated than the levels of 0.001 μCi/ml to 0.01 μCi/ml found in neutron generator pump oil by Watson et al. [8]. Less than 0.2 of 1 percent body burden exposure was received via tritium absorption during the revamping processing for any given pump as demonstrated by urinalysis.

Conclusion

The data gathered during these investigations show that considerable amounts of tritium are contained in the stainless steel body portions of ion pumps used in conjunction with Cockroft-Walton neutron generators. Depending on the history and types of targets used in these pumps, this may amount to tens of curies. The tritium from the pump interior is more soluble in H_2O by about two to three orders of magnitude, compared to outgassing by diffusion through the metal alone. Considering the health physics aspects, this would indicate that dry conditions are preferable to damp for work during reconditioning. Metallurgical studies of the peculiar coating material found inside the pump bodies suggest that titanium is sputtered throughout the vacuum system and may entrap tritium in areas other than the ion pump system.

ACKNOWLEDGMENTS

The authors wish to acknowledge the contributions of Robert Reinhart (particle sizing) and Earl Conway (microprobe analyses), both of this laboratory.

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